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The quantities of liquid transported by anomalous osmosis. I. ABRAMS and K. SOLLNER (introduced by M. B. Visscher). *Department of Physiology, University of Minnesota, Minneapolis.*

In the past, experimental studies on anomalous osmosis have been concerned solely with the anomalous osmotic pressures developed in suitable systems (J. Loeb, J. Gen. Physiol. **1**:717, 1918–19; **2**:173, 255, 387, 1919–20, etc.). If anomalous osmosis is important as a liquid moving mechanism in living systems, as many observers believe, the quantities of liquid transported may be more interesting from a biological point of view than the maximum pressures attainable. If a porous collodion membrane, activated by the method of Sollner, Abrams and Carr (J. Gen. Physiol., in print) be interposed between, e.g., 0.01 M K_3 citrate and pure water, a considerable transfer of liquid occurs. The rate of this transportation across the membrane, if the opposing pressure is small, may be as high as 90 ml. per hundred square centimeters of membrane per hour. The transport is in this case from the water to the salt solution (anomalous positive osmosis). This effect is only slightly reduced if the water is replaced by a uni-univalent salt or sugar solution of the same osmotic activity as the citrate opposite it. Even distinctly hypertonic solutions of uni-univalent salts or of non-electrolytes may be transported across the membrane, though at a lower rate. This type of flow is substantially identical with the often described phenomenon of negative osmosis—liquid movement through a membrane from the side of the concentrated to the side of the dilute solution. Anomalous osmotic phenomena do not occur with strictly semi-permeable membranes, but only with membranes of appreciable permeability. Membranes of such porosity are frequently found in biological systems.

Peripheral vascular responses in the hyperthyroid state. DAVID I. ABRAMSON and SIDNEY M. FIERST (by invitation). *May Institute for Medical Research, The Jewish Hospital, Cincinnati, O.*

The presence of a flushed, moist skin, a wide pulse pressure, an elevated skin temperature, and a decrease in circulation time in thyrotoxicosis has led to the belief that there is an increase in blood flow to the periphery in

this state. Since this view is for the most part based upon indirect evidence, the subject has been reexamined by determining the actual rate of blood flow to the extremities by means of the venous occlusion plethysmographic method.

The peripheral vascular responses were studied in 9 hyperthyroid patients, and in 5, blood flow readings were obtained before and after thyroidectomy. In each instance there was a significant increase in the rate of blood flow through the forearm, the average for the hyperthyroid group being 4.5 ± 1.05 cc. per minute per 100 cc. limb volume, as compared with an average reading of 1.8 ± 0.64 cc. for the control series. Following lugolization and thyroidectomy, there was a definite decrease in flow, with a return to the normal level within 11 to 68 days after operation. The findings in the leg were grossly similar to those in the forearm. In the hand the blood flow was not increased, the average for the hyperthyroid group being 9.7 ± 3.8 cc. per minute per 100 cc. limb volume, as compared with an average reading of 10.1 ± 3.4 cc. for the control series. Following thyroidectomy there was no significant change in 4 cases, while in 1 there was a drop from a high normal to a lower normal level.

By exposing the forearm to a controlled amount of exercise, it was found that the blood flow repayment (debt) during the hyperthyroid state was much greater than that elicited by the same procedure a short time after thyroidectomy.

On comparing the blood flow readings with the other data collected at the same time, it was noted that a gross correlation existed between the blood flow to the forearm and the basal metabolic rate, but none between the latter and the blood flow to the hand. No relationship was observed between the circulation time and the blood flow to either the hand or forearm.

The effect of bilateral paravertebral sympathectomy on the cardiorenal system in essential hypertension. WRIGHT ADAMS (by invitation), ALF S. ALVING, IRENE SANDIFORD (by invitation), K. S. GRIMSON (by invitation) and CHARLES SCOTT (by invitation). *Departments of Medicine and Surgery, University of Chicago, Chicago, Ill.*

The effect of extensive paravertebral sympathectomy on the blood pressure in patients with essential hypertension has been reported. In this paper the results of studies on the cardio-renal system before and at intervals up to seven months after operation will be presented.

The urea clearance, the ability of the kidney to concentrate urine, venous pressure, and arm to tongue circulation time showed no regular significant change after operation in either those patients who did or those who did not have a marked reduction of blood pressure.

The heart rate under basal conditions was reduced slightly after operation. There was no regular change in heart size. The vital capacity was markedly reduced in all those studied after operation. The basal cardiac output was reduced moderately after operation but the extent of the reduction was not related to the effect of the operation on blood pressure.

None of the general circulatory changes found would be expected to produce the reduction of blood pressure observed in some of the patients.

The effect of operation on the renal blood flow, gomerular filtration rate and functioning tubular mass is interesting in view of the current idea that

the lowering of blood pressure following sympathectomy might be due to the abolition of renal ischemia. One of six patients studied had no renal ischemia before operation. In one patient relaxation of the efferent arterioles occurred after operation as indicated by a diminution in the filtration fraction and increased renal blood flow. In all the other patients there occurred either a diminution of renal blood flow after operation or no significant change in blood flow. It is obvious, therefore, from these studies that such fall in blood pressure as occurs from operating on the sympathetic nervous system for hypertension is not necessarily accompanied by an increase in blood flow through the kidneys.

It seems probable that such drop in blood pressure as occurs following "total" sympathectomy is due to decrease in peripheral resistance in a large vascular bed.

Synergistic actions of drugs on the human colon. HARRY F. ADLER and A. J. ATKINSON (introduced by K. K. Jones). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Four colostomized adult males in excellent condition served as subjects for experiments dealing with the actions of several drugs in common clinical use. Colon motility was measured by the balloon technique. Sixty-seven control experiments of 3 hours duration were obtained to determine the normal colonic motility. In the drug experiments a 20 to 40 minute control period was allowed before the drug was administered.

Surgical pituitrin (Parke, Davis and Co.), administered subcutaneously or intramuscularly in doses of 1.5 to 2.5 units, within a few minutes led to the appearance of propulsive motility enduring for at least 20 minutes. Physostigmine (1 mgm.) was variable as to its onset of action and duration, but also resulted in propulsive motility. Prostigmine methylsulfate (1:4000 or 1:2000) sometimes had a variable onset of action and a variable duration of action, but was a potent agent for exciting propulsive motility. No side reactions were noted, as was sometimes the case with physostigmine. Ergotamine (0.25 mgm.) intramuscularly had no discernible action on the colon. The combination of prostigmine with either pituitrin or ergotamine was very efficacious in producing propulsive activity with expulsion of material. The additive action of prostigmine and pituitrin was marked, as was the synergistic action of prostigmine and gynergen. The simultaneous injection of 1.5 units of pituitrin, 1:4000 prostigmine and 0.25 mgm. of gynergen resulted in the appearance of motility both propulsive and non-propulsive in nature, enduring for several hours, with repeated evacuation of gas and material. Only a few experiments of this nature were attempted.

Postnatal development of water diuresis. E. F. ADOLPH. *Department of Physiology, The University of Rochester, School of Medicine, Rochester, N. Y.* (Read by title.)

Are animals provided at birth with means of promptly eliminating excesses of water? Thirteen dog pups of 5 litters were tested at various ages. They received 5 per cent of the body weight of tap water by stomach tube, were weighed at intervals, and their urine was collected whenever spontaneously voided. Results are compared according to the times required to lose diverse amounts of the administered water.

Within 10 days after birth, rate of weight loss is scarcely augmented by the presence of water excess, and little more urine is found than from the same individuals without the excess. Whereas adult dogs require 1.5 hours to eliminate half the excess, mostly in urine, pups require 3 to 5 hours, and nearly all is lost insensibly. In the second or third weeks of life prompt water diuresis is acquired, quite suddenly in each individual. At that time appear the exact temporal relations, and the urinary rates and dilutions, typical of water diuresis of the adult.

Plasma, sampled from the hearts of pups in which this function has not yet metamorphosed, is diluted (refractive index) during at least 3 hours after water administration. Sacrificed individuals show no extra fluid in stomach, intestine, or urinary bladder, and only slight transudate in peritoneal cavity. No corresponding difference of structure is detected in prepared sections of kidneys. Evidently in newborn pups the water is absorbed and distributed, but is not excreted.

In brief, water diuresis in response to water administration is perfected within a space of 3 days, at 10 to 21 days of age. If it were present before birth, it might lead to large accumulations of amniotic fluid. The function of adjusting water excesses develops postnatally the pattern characteristic of the adult dog.

Autonomic responses of bronchial tissue to various anesthetic drugs. JOHN ADRIANI and E. A. ROVENSTINE (introduced by Stevens J. Martin). *Division of Surgery, Department of Anesthesia, New York University College of Medicine, New York City.* (Read by title.)

Recently described autonomic stimulation by anesthetic drugs has aroused interest in their effects on bronchial tissue. The following experiments were performed to determine the precise role played by such drugs.

One hundred and thirteen observations with the low power microscope were made to study the size and contour of the bronchi in excised lung tissue of rats and dogs. A modified Sollman's technique was used. The preparations were immersed in solutions of anesthetic drugs dissolved in Locke's solution in concentrations approaching those in the plasma of intact animals. Photomicrographs were completed on standard film for comparison of the various responses with controls. Atropine sulphate, eserine salicylate or ergotamine tartrate solutions were added either before or after the anesthetic drug and the effects observed and compared with controls.

Cyclopropane as well as amytal, nembutal, evipal and pentothal produced a constrictor effect on bronchial musculature which could be prevented by previously adding atropine and could be relieved when atropine was added subsequently. Eserine enhanced the constrictor response. Ethylene, nitrous oxide and paraldehyde produced a mild constrictor action or no effect. The constrictor effect from these, however, was not modified by either atropine or eserine. Chloroform, ethyl ether and divinyl ether produced bronchial relaxation. Ergotamine tartrate reversed the dilator response from divinyl ether to constriction. Dilator responses persisted with ether and chloroform when preceded by ergotamine tartrate.

These experiments suggest that cyclopropane and barbiturates produce a constrictor action on the bronchial musculature by parasympathetic stimulation. Nitrous oxide, ethylene and paraldehyde constrict by a

direct action on the muscle. Ether and chloroform relax by direct action on the muscle and possibly by sympathetic stimulation. Vinyl ether exerts its action by sympathetic stimulation.

The use of the adreno-demedullated, the hypophysectomized and the hypophysectomized-adreno-demedullated rat for the assay of insulin. A. ALLEN (by invitation), J. FELDMAN (by invitation) and E. GELLHORN. *Department of Physiology, College of Medicine, University of Illinois, Chicago.*[1]

Rats fasted 18 hours were injected intraperitoneally with various quantities of insulin and the blood sugar was determined by the Somogyi modification of the Shaffer-Hartman method one hour after the injection. It was found that a fall of 10 mgm. per cent blood sugar was produced by 0.005 unit/100 gram rat in the adreno-demedullated (I), by 0.01 unit/100 grams in the hypophysectomized (II), and by 0.0003 unit/100 grams in the hypophysectomized-adreno-demedullated (III) rat. Approximately the same ratio for the sensitivity of the three groups to insulin results from the study of insulin coma or convulsions. These phenomena are produced by 0.02–0.03 unit/100 gram rat in group I and II, whereas in group III, 0.001 unit/100 grams causes convulsions or coma. The insulin assay was repeated in group III in the presence of 1 cc. of human blood/100 grams of rat. It was found that the presence of normal blood did not significantly alter the quantitative effects of insulin in group III. The enormous sensitivity of the hypophysectomized-adreno-demedullated rat makes this animal suitable for the assay of insulin in the blood (cf. abstract of paper by Gellhorn, Allen, Cortell and Feldman). Normal human blood when injected without insulin lowers the blood sugar of rats in group III by about 6 mgm. per cent. Since the injection of blood into normal rats causes no change in blood sugar (av. +0.8 mgm. per cent) the insulin content of the normal human blood is approximately 0.0001 unit of insulin per cubic centimeter.

Indirect blood pressure determinations in experiments with explanted kidneys. FREDERICK M. ALLEN and OTIS M. COPE. *Department of Physiology, New York Medical College, New York City.*

Both the indirect readings of blood pressure and the location of kidneys under the skin make possible a variety of manipulations and observations under strictly physiological conditions. In addition to the pioneer method of chronic experimental hypertension (Loesch) there are opportunities to study an acute transitory form of hypertension, also the effects of various influences, such as foods and organ extracts, upon both the blood pressure and the kidneys. The paper summarizes some results recently obtained on resumption of this work after a long interval.

The effect of pancreatic fistula on blood and liver lipids. J. GARROTT ALLEN (by invitation), CORNELIUS W. VERMEULEN (by invitation), ORMAND C. JULIAN (by invitation), DWIGHT E. CLARK (by invitation) and LESTER R. DRAGSTEDT. *Department of Surgery, The University of Chicago, Chicago, Ill.*

[1] Aided by a grant from the John and Mary R. Markle Foundation and W.P.A. Project 30278.

Depancreatized animals adequately treated with insulin commonly develop marked hypolipemia and fatty infiltration of the liver within a period of 4 to 6 weeks, depending somewhat on the amount of fat in the diet. This alteration in fat metabolism has been attributed to lipocaic deficiency. Depancreatized dogs, however, also suffer from absence of pancreatic juice in the intestine. The present study was made to determine the effect of pancreatic fistula on the blood and liver lipids of dogs. Complete external deviation of the pancreatic juice was secured in 5 animals. These survived the operation from 6 weeks to 3 months and the blood and liver lipids remained within the normal range.

The lack of inactivation of stilbestrol by the liver. M. J. ALLEN (introduced by H. Greengard). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Stilbestrol differs from the natural estrogens in that it is very potent by mouth. There is good evidence that natural estrogens are inactivated by the liver. Other workers have reported that when ovaries are transplanted intramesenterically in such a position that their venous drainage is into the portal system, they have no estrogenic effect. This liver inactivation is considered to be the reason why natural estrogens are relatively ineffective by mouth. It was considered possible that the high oral potency of stilbestrol might be due to the inability of the liver to inactivate it.

This hypothesis was tested. Stilbestrol pellets (approximately 3.0 mgm. each) were implanted intramesenterically into 10 castrate female rats and subcutaneously into 10 control castrate rats. After 48 hours the animals of both groups went into prolonged estrus. It is concluded that stilbestrol differs from natural estrogens in that stilbestrol is not inactivated by the liver. It seems probable that this fact explains the oral potency of stilbestrol.

The constitution of native activators of protyrosinase. THOMAS H. ALLEN and EDWARD BOYD (introduced by Joseph Hall Bodine). *Department of Zoology, State University of Iowa, Iowa City, and Department of Chemistry, University of Chicago, Chicago, Ill.*

Since the lipoidal phase or centripetal layer in an extract of grasshopper eggs seems to change the contained protyrosinase into tyrosinase, it should be of interest to see if there is a specific native activator of protyrosinase.

This oil and also its saponifiable fraction (mixed fatty acids) were fractionated in a sublimation apparatus at pressures less than 5×10^{-5} mm. of Hg. and between temperatures of 40 degrees to 328°C.

Surface film characteristics, the constancy of melting points on repeated fractionation, the neutral equivalents and the activation properties of the fractions were compared. As a result of these studies it seems that the oil is a mixture of activators. The latter components presumably share chemical groups of similar properties. These groups may be those polar groups of interest to surface chemistry, because a pure aliphatic hydrocarbon oil is unable to activate protyrosinase.

Comparative actions of phenyl-, thienyl- and furyl-isopropylamines. GORDON A. ALLES and GEORGE A. FEIGEN (by invitation). *Pharmacological Laboratory, University of California Medical School, San Fran-*

cisco, and Kerckhoff Biological Laboratories, California Institute of Technology, Pasadena.

Because of the physical chemical similarities of derivatives of benzene, thiophene and furan, these isopropylamines were compared in their physiological actions. The three amines produced similar circulatory effects in dogs, the intensity of the actions of the benzene and thiophene compounds being almost identical, and the furyl compound about one-third as active.

On isolated rabbit ileum, the three compounds in minimally active concentrations produce increases in tone, and with increased concentrations an effect to inhibit tone and rhythm becomes evident, most particularly with the benzene and thiophene derivatives. Dependent upon a proper molal relationship, the stimulant effects of the compounds are antagonized by atropine, and they are also antagonistic to the actions of acetylcholine and epinephrine.

The central stimulant effects, as judged by motor activities in mice, indicate the benzene derivative to be the most active of the three compounds, with thienylisopropylamine somewhat less active, and furylisopropylamine even less. Studies in man based on subjective and objective observations indicated the same order for the central stimulant effects of the compounds, but differences observed were more marked.

Effect of amphetamin sulfate on the nervous activity of dogs. E. Bryce Alpern (by invitation), Nathaniel Finkelstein (by invitation) and W. Horsley Gantt. *Pavlovian Laboratory, Phipps Clinic, Johns Hopkins University, Baltimore, Md.*

The effect of 1 mgm./kgm. amphetamin sulfate given orally was measured in 2 dogs having well established conditioned reflexes to food and in 3 dogs with stable motor defense reflexes to pain. The following effects were noted: appetite was markedly diminished, parotid secretion was slightly lessened, the heart and respiratory rates were irregular and often increased; gross muscular activity was increased or unchanged. Sexual reflexes were weakened, i.e., the onset of erection to sexual stimulation was delayed and the duration was shortened about 30 per cent. The conditioned reflexes, both motor and secretory, were less altered than were the above unconditioned reflex functions; there was little change in magnitude of the responses; the chief modification was in the direction of impairment of differentiation (inhibition often being converted into excitation); this impairment was about the same as with moderate doses of alcohol. The accompanying conditioned autonomic functions (respiration and heart beat) also suffered loss of differentiation. The latent period of the secretory conditioned reflexes was somewhat shortened, while the latent period of the motor defense conditioned reflexes was lengthened. The maximum effect of the benzedrine was roughly about 1 hour after administration but the effect could be detected for about 6 hours. Considerable individual variation was seen in the animals. Owing to the somewhat depressing effect of benzedrine on the unconditioned reflexes, there was a *relative*, though rarely an absolute, increase of the conditioned reflexes, i.e., the ratio CR/UR was greater after benzedrine than before. This is a possible explanation of the apparent stimulating effect in the human being. No loss of differentiation or conditioned reflex changes were noted on the day

following the administration of the drug. We conclude amphetamin sulfate acts in dogs as a slight depressant rather than as a stimulant to both conditioned and unconditioned reflexes but particularly the latter, regardless of what effect it may have on mood, subjective feelings, or fatigue.

Deprivation of placental blood as a cause of iron deficiency in infants. H. L. ALT (by invitation), E. E. WILSON (by invitation), Q. B. DE MARSH (by invitation) and W. F. WINDLE. *Departments of Medicine and Anatomy, Northwestern University Medical School, Chicago, Ill.*

Immediate clamping of the umbilical cord after birth deprives the infant of over 100 cc. of blood (almost one third of its total blood volume). This blood normally flows into the infant when clamping is delayed until after placental separation. We have shown previously that deprivation of placental blood leads to a decrease in erythrocytes and hemoglobin and to an increase in reticulocytes during the first week of life.

The newborn's iron reserve is principally in circulating hemoglobin. Iron liberated during blood destruction is stored in tissues and utilized as needed for hemoglobin synthesis and growth. The iron reserve is largely depleted at six months. This is attested to by the low iron content of liver and high incidence of iron deficiency anemia at this time.

The iron content of the blood left in the placenta after immediate clamping of the cord averages at least 56 mg. Failure of the infant to receive this iron results in a lower iron content throughout the nursing period. Theoretically, this loss reduces the hemoglobin from 12 grams to 9.1 grams per 100 cc. at 6 months, and from 12 grams to 9.7 grams at 9 months.

Erythrocyte counts and hemoglobin determinations were made on 28 infants, age 9 to 10 months. In fifteen infants whose cords were clamped immediately average values were: erythrocytes 5.06 ± 0.87 mil. per cmm.; hemoglobin 10.8 ± 1.9 grams per 100 cc.; mean corpuscular hemoglobin 21.8 ± 4.6 micro micrograms. In thirteen of the "delay group" values were: erythrocytes 4.45 ± 0.3; hemoglobin 11.9 ± 1.3; mean corpuscular hemoglobin 27.0 ± 4.3. Four infants in the "immediate group" had less than 10 grams of hemoglobin per 100 cc., whereas none fell below this level in the "delay group."

The decrease in mean corpuscular hemoglobin in the "immediate group" signifies a reduction in size and/or in hemoglobin concentration of erythrocytes, which is indicative of iron deficiency. It is therefore suggested that deprivation of the infant of its placental blood by early clamping of the cord may be a factor in the cause of iron deficiency anemia during the first year of life.

Studies on the salt-treated adrenalectomized rat. EVELYN ANDERSON, MICHAEL JOSEPH (by invitation) and HERBERT M. EVANS. *Institute of Experimental Biology and the Division of Medicine, University of California, Berkeley.*

Adrenalectomized rats kept under optimal living conditions and allowed to drink one per cent NaCl solution store fed glucose as liver glycogen almost as well as intact animals. They continue to grow and to maintain a satisfactory state of health. Animals so treated have been observed for periods as long as 176 days postoperative.[1] In this study such animals were fasted and the urinary nitrogen and survival time noted.

[1] Anderson, E., V. Herring and M. Joseph. Proc. Soc. Exper. Biol. and Med. **45**: 488, 1940.

Fasted adrenalectomized rats given one per cent NaCl to drink excrete nitrogen in amounts comparable with that of normal animals or adrenalectomized animals treated with desoxycorticosterone acetate, during the first five days of fasting. If tap water is given in place of NaCl solution the urinary nitrogen is reduced by 50 per cent.

Under a given set of conditions the fasting survival time of the untreated adrenalectomized rat is 4.8 days, the NaCl-treated rat 6.4 days, the desoxycorticosterone-treated rat 7 days and the sham adrenalectomized rat 13 days.

A glass capsule manometer for recording and measuring the blood pressure. FREDERICK F. ANDERSON (introduced by H. B. van Dyke). *Division of Pharmacology, The Squibb Institute for Medical Research, New Brunswick, N. J.* (Demonstration.)

The apparatus consists of a glass chamber in which a glass capsule, 80 mm. in diameter, with a capacity of about 10 cc. is enclosed. A tube containing a piston is sealed into the top of the capsule. All the glass parts are made of Pyrex glass.

The capsule is filled with distilled water and the outer chamber, connected with the artery and with a modified Trendelenburg apparatus (Pflüger's Arch. **203:** 413, 1924), is filled with isotonic saline containing 10 mgm. per cent heparin sodium. The pressure variation is transmitted to the capsule causing a change in volume and a corresponding piston-movement which is magnified by a lever writing on a kymograph. Changes in room temperature during an ordinary experiment cause no significant error of recording pressure-changes. Satisfactory tracings have been readily made in experiments with mammals and with the fowl.

Some quantitative characteristics of x-ray effects on certain cells. RUBERT S. ANDERSON, H. TURKOWITZ and K. P. LORENZ (introduced by William H. Chambers). *The Memorial Hospital, New York City.*

The fundamental mechanisms and even characteristics of the effects on cells produced by ionizing radiations, such as x-rays, are in dispute. As one step toward understanding these mechanisms, it is important to study quantitatively the sensitivity of cells under widely different conditions. In the present work, a yeast, Torula cremoris, and a bacterium, Ph. Fischeri, have been used and an x-ray effect, "delayed death" has been measured, largely by the plate count method, under several conditions.

It has been found that the presence or absence of oxygen at the time of irradiation influences the sensitivity of certain types of cultures in agreement with some earlier work. In both forms, those cultures which are influenced are less sensitive to radiation in the absence of oxygen than in its presence.

It has frequently been stated that an increased growth rate is associated with an increased sensitivity of cells. For this yeast there is no necessary correlation between increased growth rate, produced in several ways, and increased sensitivity. In fact in some cases there is an apparent effect in the opposite direction. Some but not all of this effect is eliminated when a growth method of determining the result is used instead of the plate count method.

Weight variations of muscles of adrenalectomized frogs in normal and hypotonic Ringer's solutions. C. A. ANGERER and HELENA ANGERER

(introduced by Frank A. Hartman). *Department of Physiology, The Ohio State University, Columbus.*

The leg muscles (ileofibrularis, sartorius and semitendinosus) of adrenalectomized, renal damaged and unoperated frogs (Rana pipiens) were studied for weight changes in normal and various dilutions (75 per cent, 50 per cent, 25 per cent) of Ringer's solution. For any one concentration studied, not less than 45 different frogs were employed. When the percentage increase in muscle weight for any specific concentration (e.g., 50 per cent) is plotted as a function of the immersion time (up to 8 hours and in some instance 24 hours) there is at the end of the second hour the following percentage increase over the excised weight: ileofibularis—adrenalectomized 25, renal damaged 38 and unoperated 53; as compared respectively with 26, 45 and 60 per cent for sartorius and 24, 49 and 57 per cent for semitendinosus. When any specific muscle is compared with various concentrations of Ringer's there is a progressive increase in weight with decreasing osmotic strength of the solutions employed.

The effect of oxygen tension on cyanide inhibition of the frequency of the isolated frog sinus. C. W. J. Armstrong (by invitation) and Kenneth C. Fisher. *Department of Biology, University of Toronto, Toronto, Canada.*

Isolated frog sinuses, auricles attached, were irrigated with glucose Ringer at a constant rate at 15°C. The hearts were doing no external work and the sinus frequency was followed as an indication of the rate of energy liberation in the pacemaker cells of the heart. The Ringer fluid contained 0.2 per cent glucose and 0.05 per cent sodium bicarbonate, the pH being 8.1–8.2. The conditions established permitted the maintenance of a constant frequency (average deviation less than 5 per cent) for as long as six to eight hours. Controls in Ringer were run for at least one hour and then this medium was replaced with Ringer containing a known amount of cyanide and of known oxygen tension. It was found that a cyanide concentration which just causes inhibition of the heart beat frequency at a particular oxygen tension is insufficient to inhibit the frequency at a higher oxygen tension. Hence the cyanide concentration just sufficient to inhibit the sinus frequency was investigated for a number of different oxygen tensions varying from 760 mm. to that of air.

When these cyanide concentrations are plotted against the oxygen tensions the points fall along a smooth curve indicating that the degree of cyanide inhibition of this tissue is definitely affected by the oxygen tension of the medium. It seems impossible to account for this effect on the basis of any such technical artifact as diffusion, and hence it appears to be an inherent property of the respiratory system concerned. The practical importance of such a result in connection with cyanide inhibition studies is obvious. The data suggest that there is not a simple competition between cyanide and oxygen for some enzyme such as is seen with carbon monoxide. The effect of increased oxygen tension on inhibition by azide differs qualitatively from its effect on cyanide inhibition.

The induction of estrous behavior in hypophysectomized rats. E. B. Astwood and E. W. Dempsey. *Departments of Pharmacology, Medicine and Physiology, Harvard Medical School, and the Medical Clinic of the Peter Bent Brigham Hospital, Boston, Mass.*

During the course of experiments on the induction of ovarian function

in hypophysectomized rats it was noted that certain animals exhibited estrous behavior. As there is no information on the factors which control estrous behavior in the absence of the hypophysis, it was not known whether this behavior was produced by estrogen or by corpus luteum hormone released from the ovary under the stimulus of the hypophyseal extracts, or whether the hypophyseal principles also were necessary. A study was therefore made of the effects of ovarian hormones on the sexual responses of hypophysectomized rats.

Immature and young adult female rats during the first two months following hypophysectomy were treated with a single dose of estrogen either alone or followed by progesterone. Estrous behavior was determined by observed mating responses when caged with males or by the finding of vaginal plugs or of spermatozoa in the vaginal smear. After a large dose of estrogen (16 gamma estradiol benzoate, 10 gamma estradiol, 100 gamma estrone or 1 mgm. stilbestrol) approximately 50 per cent of the animals mated within 48 hours. In the case of estradiol benzoate repeated mating occurred in a small number of animals for as long as 6 days after injection. Injections of 0.5 mgm. of progesterone into these estrogen pretreated animals was followed by mating responses within 4 hours. Graded doses of estradiol followed in 30–36 hours by 0.5 mgm. of progesterone gave results in rough proportion to the estrogen dose. This dose of progesterone given alone was ineffective, but induced mating behavior in 95 per cent of 31 animals pretreated with 10 gamma estrone, 71 per cent of animals after 5 gamma, 56 per cent of 18 animals after 2.5 gamma and 39 per cent of 18 animals after 1 gamma.

These results show that mating responses can be produced in the complete absence of the hypophyseal hormones. Sexual behavior, however, may be caused either by estrogen alone or by estrogen and progesterone. The observation of mating behavior cannot be regarded, therefore, as conclusive evidence of progesterone secretion.

Comparative studies of the respiratory act (twitch frequency and respiratory rhythm). A. K. Atkinson (by invitation) and Robert Gesell. *Department of Physiology, University of Michigan, Ann Arbor.*

The maximum twitch frequency of individual muscle units of the diaphragm was determined in the mouse, rat, rabbit, dog and horse during eupneic breathing under routine experimental conditions. This was done to learn whether or not a relation exists between twitch frequency and respiratory rhythm and the general nimbleness of action. In the mouse, which is the fastest breather and the most nimble of the group, the average maximum twitch frequency of four individuals was 84 per second. In the horse, in which frequency of breathing and nimbleness are the lowest, the maximum twitch frequency ranged between 10 and 16 per second (one experiment only). In the dog, which breathes less frequently and is obviously more clumsy than the rabbit, the average frequency was 30 per second as compared with 46 in the rabbit. The average of twitch frequency in four rats was 36 per second. Based on size alone this frequency should be higher than that of the rabbit, but sluggishness may be a determining factor. Further observations on more definitely sluggish forms such as the porcupine would help to give more positive interpretation to our findings on the rat.

It may be tentatively concluded that a short inspiration such as occurs

in the mouse allows a relatively large number of muscle fiber twitches per inspiration just as does the longer respiration of the horse where bulk and high mechanical inertia prevail. This is a most important factor for an effective gradation of the strength of contraction because the *sum total of twitches of all of the muscle units involved* (and not twitch frequency per se) is the important factor which determines the strength of muscular contraction. It is further suggested that great numerical superiority of muscle units which may be associated with great mass of muscle tissue in the horse tolerates a low frequency of twitch without endangering the evenness of the tension of muscular contraction.

Functional organization and interrelation of cerebral hemispheres in chimpanzee.[1] PERCIVAL BAILEY (by invitation), HUGH W. GAROL (by invitation) and W. S. MCCULLOCH. *Laboratory of Neurophysiology, Yale University, School of Medicine, New Haven, Conn.*

By local strychninization of one hemisphere of the chimpanzee and recording electrical activity of both, the map of functional organization has been extended and the directed functional interrelation of the two hemispheres has been explored.

The functionally unique bands have been traced in the face- and leg-subdivisions, as well as in the arm-subdivision, except that band III cannot be mapped with certainty to the medial edge of the hemisphere.

Bands I, III, VI, VII, VIII and XI have no significant projection to the opposite hemisphere. All parts of band II have such projections to contralateral band II and some parts to V, VI and VIII. Bands IV and V project across only in sectors for trunk and neck. These projections are principally to symmetrical parts. Bands IX and X have callosal projections from two regions, one above the superior parietal and the other below the interparietal sulcus.

Extension of these studies to immediately adjacent areas to control the suppressor bands I and XI disclosed contralateral connections from in front of band I and from behind band XI.

Finally, this work disclosed that strychninization of any of the suppressor bands, I, III, VII and XI, suppressed the electrical activity not only of the homolateral, but also of the contralateral hemisphere.

Observations on the localization of the Bainbridge Reflex. ROBERT BALLIN (introduced by L. N. Katz).[2] *Cardiovascular Department, Michael Reese Hospital, Chicago, and the Department of Physiology, University of Chicago, Chicago, Ill.*

Since the Bainbridge reflex was believed to be elicited through an increased venous pressure acting upon sensory endings at the junction of the superior vena cava and the right auricle, the problem was directed toward a study of these local areas. Six trained unanesthetized dogs were used to avoid depression of the reflex under anesthesia. In three other untrained animals, $\frac{1}{2}$ grain morphine sulphate was administered subcutaneously to insure relaxation.

The animals were immobilized. Heparin was injected intravenously and a special cannula was inserted into the superior vena cava through the

[1] Aided by a grant from the John and Mary R. Markle Foundation.
[2] Aided by the A. D. Nast Fund for Cardiac Research.

right external jugular vein. By means of a screw arrangement at one end of the cannula, umbrella-like ribs could be raised at the other end to distend the superior vena cava at any local area desired, without impeding venous flow.

Fluoroscopy was performed and the umbrella ribs placed into position at the junction of the superior vena cava and the right auricle. At autopsy the position was checked again, the umbrella ribs opened to check the degree of distention, and the vein examined for any possible damage. Venous and arterial pressures and heart rate were determined from Hamilton manometer records.

Repeated distension of the superior vena cava produced no significant change in the heart rate or the arterial blood pressure. However, injection of 200 cc. of the Ringer's solution over periods of from 23–38 seconds caused a significant increase in heart rate as well as in the arterial pressure. The heart rate returned to normal in 3–8 minutes. In three untrained dogs which were morphinized, repeated distension of the superior vena cava was accomplished without change in either heart rate or arterial pressure; increasing the venous return produced the usual cardiac acceleration.

These results indicate that the sensory receptors eliciting the Bainbridge reflex in unanesthetized dogs lie elsewhere than in the nerve endings in this part of the superior vena cava.

The narcotic action of CO_2 in the albino rat. J. H. Barbour (by invitation) and M. H. Seevers. *Department of Pharmacology, University of Wisconsin, Madison.* (Read by title.)

When the albino rat is exposed acutely to CO_2 the maximum tolerated concentration lies between 15 and 20 per cent. At 20 per cent an occasional animal will survive. Atmospheres containing between 25 and 50 per cent CO_2 are uniformly lethal, survival time varying between one-half and thirty-six hours, depending on the concentration of this gas. At 50 per cent CO_2, no animal survives longer than five hours. Death seems to be due primarily to pulmonary injury rather than narcosis. The edema and hemorrhage noted in the lungs are also seen in all exposed mucous membranes.

An immediate narcotic action is observed at all concentrations above 30 per cent CO_2. Narcosis is less pronounced at lower levels (20 to 30 per cent CO_2) and is not detected at 10 per cent CO_2.

That depression of certain functions does occur at 10 per cent CO_2 is indicated by a temporary decrease in the oxygen consumption of 10 to 25 per cent below the basal level at 28°C. during the first two to four hours of exposure. Thereafter, a gradual return to the basal level occurs, the time required varying with the individual, but usually complete before twenty-four hours. These changes in oxygen consumption may be related to variations in the narcotic action of CO_2 and cold, described elsewhere in these abstracts.

The maximum tolerated concentration of CO_2 during chronic exposure lies between 20 and 25 per cent. If such concentrations are attained gradually during 5 days, they are tolerated for 30 days or more, whereas sudden exposure is lethal within 2 to 5 days. Narcosis is not a prominent feature if the concentration of CO_2 is held below 25 per cent, although

food consumption is greatly decreased and 50 per cent of the body weight is lost. On removal to air, irritability and tetany are observed for 12 to 18 hours followed by complete recovery. During thirty days exposure to 10 per cent CO_2, no depression is noted although a steady loss in weight occurs (14 to 25 per cent). Young, born during exposure, develop normally.

No tolerance to the respiratory action of CO_2 has been detected in any of these experiments involving chronic exposure.

The influence of cold on the narcotic action of CO_2. J. H. BARBOUR (by invitation) and M. H. SEEVERS. *Department of Pharmacology, University of Wisconsin, Madison.*

A narcotic state having some characteristics of both anesthesia and hibernation is induced in rats, rabbits and dogs by exposure to CO_2 in a chamber at 5°C. The minimal narcotic concentrations of CO_2 at this temperature are, rat 11 per cent, rabbit 17 per cent, dog 14 per cent, the induction time varying from 3 to 24 hours. These concentrations of CO_2 do not induce narcosis at 25°C.

The following observations apply to the rat 3 to 6 hours after exposure to 11 per cent CO_2 at 5°C.: loss of most reflexes; hypothermia 16 to 21°C. (rectal—thermometer insertion 50 to 75 mm.); bradycardia with arrhythmia (rate 30 to 100 per min.); and bradypnea (rate 2 to 20 per min.). Once narcosis is established, the environmental temperature becomes the principal factor modifying the subsequent course of events. At 5°C., narcosis progresses rapidly even in air, becoming irreversible when the body temperature falls to 13°C. At 19°C. (CO_2 11 per cent), the level of narcosis remains fairly constant. Recovery follows removal prior to 4 hours, but longer exposure (6 to 12 hrs.) is fatal. At 25°C. (11 per cent CO_2 or air), reflex activity is regained completely within 2 to 3 hours. The induction time is reduced significantly by a 24-hour fast or by previous exposure to 10 per cent O_2 for three weeks. Weight, sex, and dehydration (24 hrs.) do not modify induction time significantly. Some resistance (prolongation of induction time) is acquired by repeated narcotizations at 3 to 4 day intervals.

The capacity of the rat for adaptation is remarkable since complete resistance to the narcotic action of 11 per cent CO_2 at 5°C. is obtained either by previous exposure to 11 per cent CO_2 at 25°C. for 24 hours, or to air at 5°C. for one week.

A similar narcotic state is induced in some rats by exposure to 5 per cent CO_2 and 10 per cent O_2 at 5°C. Exposure for 36 to 48 hours to 10 per cent O_2 at 5°C. results in narcosis which is dissimilar to that induced by CO_2 at 5°C.

Organic phosphate changes in resting cardiac muscle as indicated by radioactivity studies.[1] S. B. BARKER, R. F. FURCHGOTT (by invitation) and EPHRAIM SHORR. *The New York Hospital, and the Department of Medicine, Cornell University Medical College, New York City.*

Radioactive phosphorus in the form of sodium phosphate[2] has been

[1] Aided by grants from the Committee on Research in Endocrinology of The National Research Council and from the Carnegie Corporation.

[2] The radioactive phosphorus was generously supplied by Dr. J. H. Lawrence of the Radiation Laboratory, University of California, Berkeley.

used to follow the uptake of phosphate in resting cardiac muscle. Slices of heart muscle from well-fed dogs were first given a 30 minute period in oxygen in a Ringer-phosphate-glucose medium, and then were exposed for periods of 10, 20, 30, and 40 minutes to a similar solution containing radioactive phosphate. Aliquots of tissue taken at these time intervals were analyzed for acid-soluble phosphate: fractions representing inorganic, phosphocreatine (PC), adenylpyrophosphate (APP), and hexose monophosphate (HMP) phosphorus were isolated for parallel measurements of activity and of phosphate. Respiration studies were run concomitantly to determine the actual extent of carbohydrate oxidation under these experimental conditions.

Exchange between tissue inorganic phosphate and that in the medium was rapid and approximately equal at 27.5°, 37.5°, and 41.0°. The activity of the inorganic phosphate in the tissue rose to about one-third of the value in the external medium. At each different temperature, the phosphorus turnover from 10 to 40 minutes was approximately equal in the PC and APP fractions and considerably lower in the HMP. After 40 minutes' incubation at 37.5°, the activities of the PC and APP fractions were about one-half of that of the tissue inorganic phosphorus, while the HMP was one-tenth. At 37.5°, oxygen consumption was 50 per cent greater than at 27.5° and there was observed increased activity in the PC, APP, and HMP fractions of the same order. There is an increase in oxygen consumption of 15–30 per cent when the temperature is elevated from 37.5° to 41.0°, paralleled by a corresponding increase in the phosphorus turnover of the fractions studied.

Some factors in the control of the human appetite. Broda O. Barnes and Robert W. Keeton. *Department of Medicine, University of Illinois, Chicago.*

Although everyone agrees that weight reduction depends primarily on a restriction in calories, many writers have insisted that better results were obtained by the injection two or three times a week of some of the endocrine preparations. The present study is concerned with the influence of these extracts on weight reduction and some of the factors affecting the human appetite.

Patients weighing between 200 and 400 pounds were hospitalized continously over periods of three to five months. They were allowed to select any type of food and in any quantity which their appetites dictated. Candy, pie, cake, or other desserts were allowed *ad libitum*. After selecting their food, a special dietician weighed the quantity and calculated the caloric value. After a control period of observation, therapy was instituted.

Injections consisting of placebos, Polyansyn (furnished by Armour and Company), or posterior lobe extracts were given two or three times weekly or in some cases daily. Most of the patients lost weight during the control period. The injection of sterile saline or either of the pituitary preparations had no influence on weight loss of any of the patients nor on the number of calories selected in the diet. It would appear that frequent injection of pituitary preparations acts largely by focusing the patients' attention on their problem. Similar results were obtained by hospitalization in those cases not previously treated.

Recently (at the suggestion of Dr. R. B. Oesting) we have been using a weight reduction diet containing about 1300 calories, which includes 51 grams of carbohydrate, 70 grams of protein, and 89 grams of fat. This diet differs from the usual reduction diet in containing less carbohydrate and more fat. Apparently some patients are more comfortable on it than on a diet low in fat.

Acetylcholine as the cause of the "negative variation" in nerve. T. CUNLIFFE BARNES (by invitation) and R. BEUTNER. *Department of Pharmacology, Hahnemann Medical College and Hospital of Philadelphia, Philadelphia, Pa.*

Acetylcholine is known to be associated with nervous activity (Loewi; Dale). Originally it was found at the nerve endings, but more recently all along the nerve fibers as well. What possible importance acetylcholine may have in the propagation of nerve impulse along the fibers is not yet known. We have now performed model experiments with "oil cells" which demonstrate the production of a negative electrical potential by the mere contact of an extremely dilute acetylcholine solution with various water insoluble substances resembling lipoids.

This finding is not surprising in view of the fact that solutions of the hydrochlorides of all organic bases, particularly alkaloidal salts, produce a marked electrical negativity when in contact with oil or lipoids (Beutner: J. Amer. Chem. Soc. **36:** 2045, 1914; Physical chemistry of living tissue, 1933).

The electrical negativity following acetylcholine is, however, outstanding by its size, its rapidity of appearance on application and disappearance after removal, and the extremely low concentration of acetocholine required.

All of these observations support the assumption that acetylcholine is of vital importance for origin of the negative electrical wave. Lillie's local circuit theory in turn explains the importance of the negative electric wave for the propagation of the nerve impulse; hence *acetylcholine might well be a factor in the propagation of the nerve impulse.*

In this model of electrical phenomena in nerve, mecholyl (acetyl-beta-methylcholine chloride) was used, owing to its greater stability. The oil layer (guaiacol or nitrobenzene or various other oil substances) made contact on each side with 0.7 per cent NaCl connected by salt bridges to beakers containing 0.7 per cent NaCl into which dipped Ag-$AgCl_2$ electrodes leading to the E.M.F. terminals of a Leeds and Northrop thermionic amplifier (for high resistance circuits) acting as a null instrument for a potentiometer. At the "oil" layer 0.000076 per cent mecholyl produced a potential of 0.5 mv., negative on the side to which the alkaloid was added. The potential was a linear function of the ion concentration of mecholyl up to about 0.003 per cent (14 mv.); at higher concentration the difference of potential followed the logarithm of the concentration. The highest potential obtained was 200 mv. (negativity) with 0.03 per cent mecholyl and nitrobenzene.

Changes in the impedance properties of adjacent body segments during intravenous injection of isotonic solutions. A. BARNETT (introduced by S. E. Barrera). *Department of Psychiatry, New York State Psychiatric Institute and Hospital, New York City.* (Read by title.)

It is known that the bulk distribution of tissue fluids shifts frequently in the body under the effect of physiological changes. It becomes important, therefore, to develop methods for following these tissue fluid shifts in individual body segments. It has been shown (Proc. Soc. Exper. Biol. and Med. **44:** 142, 1940) that when normal saline or Ringer's solution is injected intravenously in man, the consequent increase in interstitial fluid causes the a.c. resistance of the arm-to-arm segment to decrease by about 10 per cent for each liter of saline injected and retained. In order to follow the course of hydration in adjacent body segments, the a.c. resistance and Q-factor (1) of a 12–14 cm. length of the upper arm and (2) of the adjacent chest segment were measured at 5–10 minute intervals during, and for ½ hour to 1 hour after, the intravenous injection of one liter of isotonic Ringer's solution which required one hour (Proc. Soc. Exper. Biol. and Med. **30:** 543, 1937). Female subjects were used because their resistances are higher (West. J. Surg. **45:** 380, 1937) and the absolute changes greater. Decreases in resistance and Q-factor were observed in the chest segment before they could be detected in the arm. In certain individuals, the electrical values first increase in the arm while decreasing in the chest, then, as the injection continues, both arm and chest values follow a decreasing course together. In other individuals, the chest values decrease practically continuously as injection progresses, the arm values varying very little until all of the Ringer's has been injected, whereupon a decrease in the arm values begins which continues for a half hour or more after injection. In general, it may be said that the chest segment responds more rapidly and to a greater extent than the arm segment to which it is attached.

Electrically produced flicker in darkness. A. BARNETT (introduced by S. E. Barrera). *Department of Psychiatry, New York State Psychiatric Institute and Hospital, New York City.* (Read by title.)

When alternating current of low intensity and frequency (⅓ cycle to 35 cycles per second) and of sine form is fed transversely across the human head, a subjective sensation of light flicker may be made to appear varying in frequency with that of the current. Non-polarizable silver-silver chloride electrode wrapped in saline soaked absorbent cotton are used (one inch square) and are mounted on the temples or in contact with saline pads inserted into the external auditory canal. Current values of 0.2–0.8 milliampere are usually sufficient to produce the effect. With the subject in darkness, if the current be gradually increased at a series of fixed frequencies and the thresholds for the first appearance of flicker be taken, the curves of current strength against log frequency plotted from the results (Cold Spring Harbor Symp. **4:** 150, 1936) takes the form of three intersecting U-shaped curves with minima at 22 cycles, 7 cycles and 4.5 cycles. It has been shown that the 22 cycle minimum may be obtained in bright light and the 7 cycle minimum in dim light (J. Psych., in press). However, it was not certain whether these minima represented intrinsic electrical response constants of the optic tracts or whether they were characteristics of the latter as modified by the effect of light falling on the retina. Since these same minima are obtained in total darkness, it may be concluded that 7 cycles is the optimum frequency for electrical stimulation of those portions of the optic tracts connected to the rods and

22 cycles the corresponding optimum frequency for those portions connected to the cones and that neither of these optima is affected by light falling on the retina. The significance of the 4.5 cycle optimum is not known. It may be associated with the mechanisms responsible for idioretinal light.

Electroencephalographic findings associated with electric shock therapy in patients with mental disorder. S. EUGENE BARRERA and BERNARD L. PACELLA (by invitation). *Department of Psychiatry, New York State Psychiatric Institute and Hospital, New York City.*

Using a two stage ink-writing recorder, push-pull amplifier system, with bi-polar readings, records were obtained during electric shock therapy on a series of patients suffering from mental disorders of various types. Records were obtained before administration of any treatment, during the course of single treatments, and at various intervals during the course of therapy and subsequent to the cessation of therapy.

The observations may be divided into two main groups; (I) Those associated with individual shocks resulting in *a*, minor or petit mal seizures, or *b*, generalized or major seizures; and (II) those associated with administration of successive shocks and with the effects remaining after cessation of the course of therapy. The essential "pathological" feature associated with the electric shock therapy in general was found to be the occurrence of slow potentials of a frequency of 3–6 cycles per second occurring at random, or frequently in serial bursts, and which may persist for a considerable period of time following the cessation of the course of treatment. The incidence of these potentials seems to be related to the number of convulsive seizures. There is no definite relationship between the therapeutic status of the patient as a whole and the incidence and persistence of the slow potentials following cessation of treatment. Electroencephalographic manifestations associated with the administration of only the petit mal type of response were relatively "mild" and transient as compared to those associated with the administration of major seizures.

Some evidence in support of a sulfhydryl mechanism of blood clotting. J. PERCY BAUMBERGER. *Physiology Department, Stanford University, Calif.*

Fibrinogen may be converted to an incoagulable insoluble form by the photodynamic action of methylene blue in the presence of oxygen. The oxygen consumption during this process has been determined by means of manometric and polarographic methods. The results show that as the photodynamic oxidation proceeds, successive samples of fibrinogen clot less and less rapidly on addition of thrombin or serum until finally no clot is obtained. When four molecules of oxygen have been consumed per molecule of fibrinogen, both the process of clotting and of photodynamic oxygen consumption cease. This stoichiometric ratio is in fair agreement with the cysteine content predicted by Bergmann.

Whether or not S-S or SH groups occur in the fibrinogen molecule was studied polarographically. Using the method of Brdička, fibrinogen gives the two polarographic waves characteristic of proteins containing cystine or cysteine or both.

Iodoacetate added to fibrinogen produced no decrease in the height of

the polarographic "protein waves." However, when treated with a detergent, Duponal WA, by Anson's method, the polarographic "protein waves" of fibrinogen can be suppressed by monoiodoacetate. This shows that most of the thiol groups are in the reduced form but are only readily accessible after denaturation.

When the thiol groups of fibrinogen are in the SH form clotting can occur but if any of these are oxidized by photodynamic action, clotting is delayed and the clot is less firm, and if all thiol groups are oxidized, no clotting results from the addition of thrombin. Bernheim cites literature indicating that thrombin is only active in the S-S form and suggests that naphthoquinone may insure the presence of the S-S form in thrombin. Kühnau and Morgenstern obtain a faint nitroprusside reaction with purified thrombin. This would indicate that the thiol groups are more accessible in thrombin than in fibrinogen and might be oxidized by naphthoquinone leaving fibrinogen SH unaffected.

We propose the hypothesis that the long crystals of fibrin that make up a clot are the result of the lining up of molecules through the formation of bridging S-S groups produced by oxidation of the SH groups of fibrinogen by thrombin.

Circulatory changes resulting from the inverted posture in normal and shocked animals. J. D. Beale, Jr. (by invitation), L. L. Chastain (by invitation) and H. S. Wells. *Department of Physiology and Pharmacology, School of Medical Sciences, Wake Forest College, Wake Forest, N. C.*

In a series of 33 dogs, anesthetized with barbital, inversion at an angle of 70° almost invariably elevated arterial pressure, provided the head was ventro-flexed. Maximum elevations often required 30 minutes for full development and ranged from 2 to 64 mm. Hg, the mean being 25 ± 11.4 mm. There was no correlation between the prevailing levels of pressure, which ranged from 45 to 180 mm. Hg, and the elevations resulting from inversion, except that a minimal change was more apt to occur above 150 mm. In shock from hemorrhage or trauma, when blood pressure was maintained below 70 for many hours, inversion resulted in the usual elevation of pressure unless vasomotor, respiratory or cardiac failure had meanwhile developed to a severe degree. In our opinion these terminal states can be relieved or postponed if shocked animals are kept in the inverted posture.

In 12 of 21 observations on 16 dogs inversion produced no significant change in cardiac output; in 7 cases it increased 18, 22, 26, 34, 37, 63 and 96 per cent; while in 2 instances it decreased 27 and 28 per cent. Oxygen consumption increased 7 to 71 per cent in 6 of the 7 cases showing increased cardiac output. Respiratory rate and volume were correspondingly increased, a change which, in variable degree, commonly resulted from inversion.

Inversion elevated intrathoracic pressure 1 to 3 cm. water, but caval venous pressure at heart level was usually increased more, so that cardiac filling pressure was probably increased.

It is concluded, tentatively, that inversion produces a primary, gravitational elevation of arterial pressure and that this immediate rise accelerates the coronary and medullary circulations to produce progressive

improvement of vasomotor tone and/or cardiac efficiency, so that blood pressure continues to rise for some time. Other factors doubtless complicate the picture.

Oxygen poisoning of unicellular organisms and its relation to mammalian tissues. JOHN W. BEAN. *Department of Physiology, University of Michigan, Ann Arbor.* (Read by title.)

There is evidence which suggests that the direct toxic action of oxygen at high pressures on isolated tissue is mediated through a poisoning of the same link in the chain of respiratory enzyme processes as that affected in cyanide poisoning. If this direct toxicity of oxygen at high pressure is due purely to a cyanimimetic action—pneumococcus, which is not appreciably affected by cyanide, should not be altered by oxygen at high pressure. In order to get experimental evidence bearing on this point, pneumococcus type I was exposed to various pressures of oxygen in a pressure chamber the temperature, humidity and CO_2 content of which were maintained constant. It was found that organisms seeded on blood agar plates failed to show any growth at oxygen pressures just greater than 900 mm. Hg. Some few of these organisms, however, remained viable after 24 hours exposure to this pressure as they did also to pressures as high as 2660 mm. Hg as shown by a definite but scant growth when subsequently exposed to control conditions for 36 hours. Oxygen pressures of 4600 mm. Hg not only completely inhibited all growth during the period of exposure to this pressure but must have killed all the organisms since none showed growth when subsequently returned to control conditions. The growth of pneumococcus type I is not appreciably affected by air pressures as high as 4600 mm. Hg. It may be concluded therefore that oxygen at just over 900 mm. Hg pressure completely inhibits growth of pneumococci and at 4600 mm. Hg pressure **not** only inhibits growth but kills these organisms. This action is not due to pressure *per se*; while acute oxygen poisoning may involve the same link in the respiratory enzyme processes in mammalian tissue cells as that affected by cyanide, its action is not limited to this specific point but rather attacks respiratory enzymes on a wider front.

Alteration in the conductivity in the mammalian heart induced by oxygen at high pressure. JOHN W. BEAN and W. V. WHITEHORN (by invitation). *Department of Physiology, University of Michigan, Ann Arbor.* (Read by title.)

In oxygen poisoning in dogs, there commonly occur cardiac changes, particularly a bradycardia upon which a tachycardia may be superimposed during convulsive seizures. In the excised frog heart also, there is a similar slowing of heart rate as a result of a direct influence of oxygen at high pressure on the pace setter mechanism. One might reasonably suppose there might be equally pronounced changes in conductivity of the cardiac impulse under these conditions. To examine this possibility the electrocardiograms of dogs decerebrated under transient ether anesthesia; tied back down in a pressure chamber; and exposed to oxygen pressures of 5 atmospheres, were recorded. Either the three standard leads with subcutaneous contacts, or an esophageal-rectal lead, were employed. The latter proved advantageous in minimizing the possible

involvement of the electrical changes in the somatic muscles during convulsive seizures. In dogs with vagi intact the heart was slowed and the P-R interval reversibly prolonged by as much as 40 per cent. The degree of recovery was dependent upon individual differences in susceptibility and the length of the exposure. In vagotomized animals there was no significant change either in the consequent tachycardia or in the P-R interval.

These results indicate that the early bradycardia of oxygen poisoning is dependent largely upon vagal integrity and that the initial prolongation of the P-R interval recorded in non-vagotomized animals exposed to oxygen at high pressure is of vagal origin. The terminal slowing in the heart rate and the accompanying prolongation of the P-R interval is not, however, dependent upon vagal integrity since it occurred in both vagotomized and non-vagotomized animals exposed to high oxygen pressure.

Localization of the medullary respiratory centers in the monkey. LINDSAY E. BEATON (by invitation) and H. W. MAGOUN. *Institute of Neurology, Northwestern University Medical School, Chicago, Ill.*

Employing the Horsley-Clarke technic, 14 monkeys have been subjected to systematic exploration of the medulla with thyratron regulated condenser discharges at a frequency of 300/sec. and intensities from 1 to 8 volts. It has proved possible to delineate two regions from which, respectively, sustained inspiratory and sustained expiratory responses can consistently be elicited. Such responses consist of a cessation of rhythmic respiration with the chest and diaphragm fixed in a position of increased inspiration or expiration.

The inspiratory area lies dorsally to the rostral half of the inferior olivary nucleus. At its anterior extremity the inspiratory field is found close to the dorsal border of the inferior olive, extending from the midline 4 mm. to either side. Near its caudal extremity this center occupies a position more extensive both ventrally and dorsally, reaching from a point between the two inferior olives to one immediately beneath the hypoglossal nucleus. At this posterior level, however, the region from which inspiratory responses are invariably obtained extends only some 2.5 mm. from the midline.

The expiratory area surrounds the inspiratory, lying rostrally, laterally and caudally to the latter, and also dorsally to it except at that level where the inspiratory field reaches the hypoglossal nucleus. In addition, scattered expirations are elicited on stimulation beneath the inspiratory center. Reaching the midline only ahead of the inspiratory region, the expiratory center extends laterally up to 7 mm. from the median raphe. It is, for purposes of rough visualization, coextensive with the reticular formation from 1 mm. rostral to the inferior olive to the latter's caudal end, excepting the compact inspiratory center. Other medullary structures are not responsive.

The anatomical location of the respiratory centers in the monkey coincide in general with those previously outlined for the cat. The inspiratory field is less and the expiratory field more disperse in the monkey than in the cat. The variations in the topography of the excitable regions in the two animals do not seem too great to be explained by changes in the arrangement of medullary structures in members of different phylogenetic orders.

On the phosphorylation hypothesis of glucose absorption, with special reference to phlorizin. LYLE VIBERT BECK (introduced by J. F. McClendon). *Department of Physiology, Hahnemann Medical College, Philadelphia, Pa.*

The finding by Kalckar (Enzymologia **2:** 47, 1937) that low concentrations of phlorizin inhibit the phosphorylation of hexoses by rabbit kidney brei has been confirmed. Phlorizin also appreciably inhibits the acid phosphatases of rat kidney cortex and intestinal mucosa, while having little effect on the corresponding alkaline phosphatases.

Combined barium precipitation—n. HCl hydrolysis experiments, by procedures suggested by the work of Lohmann, indicate that the increase in organic phosphate produced in the intestinal mucosa of rats by giving them glucose by stomach tube (cf. Laszt and Sullmann, Biochem. Ztschr. **278:** 401, 1935) is due to increase in concentration of several organic phosphate compounds. The considerable increase in the 0–7 minute n. HCl fraction is apparently largely due to formation of adenylpyrophosphate, since this fraction was found mainly in the Ba precipitate fraction. Increases in the 7–180 minute n. HCl fraction and in the difficultly hydrolyzable fraction (not hydrolyzed in 180 minutes by n. HCl at 100°C.) are due chiefly to formation of carbohydrate-phosphate compounds, since these fractions were found mainly in the Barium soluble fraction.

These experiments indicated that in the mucosa there is almost no glucose-1-PO_4. Formation of hexose-6-PO_4 on giving glucose is indicated by the increase in Ba soluble, difficultly hydrolyzable organic phosphate. Also, under these same conditions an increase in concentration of compounds giving the Seliwanoff fructose reaction (Roe. J. Biol. Chem. **107:** 15, 1934) occurred.

M/50 phlorizin did not affect the increase in the 0–7 minute n. HCl (pyrophosphate) fraction produced by 5.5 per cent glucose, but definitely diminished the increases otherwise produced in the 7–180 minute fraction and in the difficultly hydrolyzible fraction. The increase in concentration of compounds giving the fructose reaction when glucose was given was diminished in the presence of m/50 phlorizin.

While the above findings may be interpreted as indicating that phosphorylation-dephosphorylation processes play a considerable rôle in glucose absorption, they suggest that if this is true the pathways involved are considerably more intricate than a simple phosphorylation and dephosphorylation of the glucose molecule.

Correlation of prenatal apnea, intrauterine respiratory movements, and respiration at birth with lung structure. R. F. BECKER (by invitation), W. H. WHITEHEAD (by invitation) and W. F. WINDLE. *Department of Anatomy, Northwestern University Medical School, Chicago, Ill.*

It has been held by some that intrauterine respiratory movements occur normally in the human fetus near term and that the consequent aspiration of amniotic fluid may serve a useful function. The theory finds no support in experiments in the guinea pig. Histologically the full term human fetal lung usually shows evidence of aspiration, but it is possible to obtain material only after death in utero; asphyxia as a cause of the aspiration can not be excluded.

We have been able to observe no intrauterine respiratory movements in the unanesthetized, unasphyxiated, pregnant guinea pig near term. Direct observation of fetuses through the thin walled gravid uterus exposed under a local anesthetic revealed no fetal respiratory movements. When an apneic fetus whose trachea had been clamped in utero was delivered, preserved in formalin and later prepared histologically, the lungs presented a compact gland-like appearance; bronchi and bronchioles possessed definite lumens but more distal passages and alveoli were collapsed. The lungs were less open than those of the usual human stillborn.

Asphyxia induced experimentally led to an increase in fetal activities; rapid but shallow respiration-like rhythms of movements sometimes appeared. As the asphyxia became more marked the fetuses began to execute true respiratory movements in utero; these were slow and deep, and aspiration of amniotic fluid was clearly observed. Sections of lungs of fetuses aspirating in utero under asphyxial conditions showed varying degrees of expansion of respiratory bronchioles and alveolar ducts.

Unasphyxiated fetuses delivered by Cesarean section under local anesthesia as well as by normal labor began to breathe almost immediately. After breathing for five minutes the lungs showed fully expanded respiratory bronchioles and alveolar ducts but most alveoli were still collapsed. During the course of the first postnatal day expansion of the newborn lung increased progressively.

Respiration of the midgut gland of the Kelp crab (*Pugettia producta*) **in relation to body size.** H. S. Belding (by invitation), J. Field, 2d and F. W. Weymouth. *Laboratory of Physiology, Hopkins Marine Station, Stanford University, California, and Department of Zoology, University of Connecticut.*

Relatively few data are available concerning the metabolism of marine invertebrate tissue *in vitro*. The present investigation, which is part of a general study of metabolism in the marine invertebrates of Monterey Bay, California, serves to supply additional data of this sort. A series of 61 crabs, 28 males and 33 females, of graded size, was studied. Oxygen consumption of excised midgut gland was measured by the Warburg manometric method.

In this series body weight ranged from 0.50 to 275 gm. and carapace length (rostral notch to posterior margin of carapace) from 1.30 to 9.68 cm. Q_{O_2} (in c.mm. oxygen, N.P.T., per mgm. dry weight per hour) ranged from 0.47 to 3.98 (15°C.). It was found that the regression lines of Q_{O_2} on both carapace length and log body weight were linear. The coefficient of correlation between Q_{O_2} and carapace length was -0.845 for the whole series (there was no significant difference between the sexes) and that between Q_{O_2} and log body weight was -0.867. The corresponding standard errors of these correlations were 0.0369 and 0.0320; the corresponding standard errors of estimate were 0.434 and 0.405.

Since the correlation between body size and age is close in these animals (Weymouth, McMillin and Rich, 1925), it may be assumed that a decrease in the Q_{O_2} of the midgut gland occurs with increase in age. This relationship is similar to that shown by various workers for certain mammalian organs (Pearce, 1936).

Concentrations of ascorbic acid and the phosphatases in secretions of the male genital tract. OWEN C. BERG (by invitation), CHARLES HUGGINS and CLARENCE V. HODGES (by invitation). *Department of Surgery, The University of Chicago, Chicago, Ill.*

In dogs with surgical isolation of the prostate gland permitting the collection of prostatic fluid for many months, the resting semen occurring without adventitious stimulation was compared with semen obtained following intravenous injection of pilocarpine. Resting semen is consistently slightly more alkaline (0.6 pH units), contains slightly less total CO_2 (0.8 mM), and considerably less chloride (52 m.-eq. per liter) than stimulated semen. A reciprocal relationship was found between the phosphatases in resting and stimulated semen; in resting semen alkaline-phosphatase and acid-phosphatase are respectively high and low in concentration, while stimulation with pilocarpine causes these values to be reversed.

It has previously been shown that the concentration of ascorbic acid is considerably higher in semen than in the plasma of guinea pigs. Ascorbic acid was found at the plasma level in prostatic fluid of guinea pig, dog and man, whereas in the seminal vesicle fluid of man and guinea pig it was considerably increased. The ascorbic acid concentration of human ejaculate was considerably higher than that of human resting seminal vesicle fluid. The seminal vesicle is thus the chief source of ascorbic acid in semen, and its concentration increases during active secretion by this structure.

The regulation of cholic acid output in the acholecystate state. A. L. BERMAN and E. F. SNAPP (introduced by F. T. Jung). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

We have studied the regulation of cholic acid output in "suction" biliary-duodenal fistula dogs when the bile is circulated through the entero-hepatic circuit hourly day and night. In this experiment we have attempted to approximate as closely as possible the situation in the cholecystectomized patient whose bile is constantly being re-circulated through the entero-hepatic circuit because of the absence of the storage and concentrating functions of the gall bladder. After the animal reached its basal cholic acid output for our standard diet with no return of bile, the bile secreted during each hour, except for 0.1 cc. used for cholic acid determination, was returned to the duodenum during the 5 to 10 minutes of the succeeding hour so that the cholates made 24 entero-hepatic circuits each day. During the experiment the standard diet was fed in divided amounts three times a day, thus keeping the basal cholate synthesis constant. The cholic acid output increased rapidly until the 40th hour, when a constant level of 0.53 gram per hour or approximately 4.3 grams per 8 hours was attained. When the cholates are circulated three times in 24 hours, the homeostatic level of 4.1 grams per 8 hour period was reached at approximately the 240th hour. When we analyzed our results by fitting them to the equation for a straight line, we obtained, for example, the following results during a typical experiment: percent recovery = 89 per cent (actual, 93 per cent; basal cholate synthesis = 0.50 gram per 8 hours (actual, 0.54 gram; homeostatic level = 3.9 grams per 8 hour period (actual, 4.3 grams). Thus it appears that regardless of the number of

entero-hepatic circuits, i.e., within the limits studied by us (3–24 per day), the regulatory mechanism for cholic acid output seems to be the same, except in reference to the speed in reaching the optimum output level. The homeostatic level depends on the basal cholic acid synthesis which in turn depends on the amount of protein in the diet.

Reversals of blood pressure responses by changing frequency of forebrain stimulation. CHARLES BERRY and ROBERT HODES (introduced by S. W. Ranson). *Institute of Neurology, Northwestern University Medical School, Chicago, Ill.*

Electrical stimuli consisting of short duration pulses of constant form at frequencies controlled between 1 and 300 per second were delivered through bipolar electrodes oriented by a Horseley-Clarke instrument. Cats were used and the cardiovascular responses were simultaneously recorded on cathode ray oscillograms from the left inferior cardiac nerve and on kymograms of carotid blood pressure.

Throughout most of the hypothalamus proper, stimuli at rates of 5 or more per second usually caused rises in blood pressure associated with increases in electrically detectable activity in the cardiac nerve. Stimulation of these same points with rates below 5 per second usually changed the responses to rapid falls providing the shocks were of considerable intensity, 10 or more peak volts. During the depressions, each stimulus was followed by a simple or complicated spike; between such spikes, the sympathetic activity was reduced or abolished in the electrical records from the cardiac nerve.

From areas surrounding the hypothalamus anteriorly (preoptic regions), dorsally (thalamus), laterally (internal capsule), and ventrally at the surface of the brain, depressor responses were frequently obtained with a wide range of frequencies up to 300 per second. During these depressions, the general activity in the cardiac nerve was depressed with no spikes driven by the stimuli. At only a few points in these regions surrounding the hypothalamus could the depressors be converted to pressors as the stimulus rate was increased. These conversions occurred at various unpredictable critical frequencies between 20 and 150 per second, but were obtainable with stimuli maintained below 10 volts.

Thus, two types of blood pressure reversals with changes of stimulus rate were revealed by differences in their ranges of critical frequencies, in their associated electrically detected sympathetic outflow, and in their localization in the forebrain.

Studies on the etiology of traumatic shock. C. H. BEST and D. Y. SOLANDT. *Departments of Physiology and Physiological Hygiene, University of Toronto, Toronto, Canada.*

Traumatic shock has been produced in a large number of dogs. Fluid loss at and adjacent to the site of injury was measured by a volume-displacement technique. From these experiments we conclude that, although many animals in so-called traumatic shock die as a result of local fluid loss, in the majority of cases this is not the only etiological factor. A type of traumatic shock can be produced in which this fluid loss, although considerable, is not alone great enough to cause death. Local fluid loss is, as Blalock has repeatedly emphasized, a major factor in the production of

such shock, but some additional etiological factor or factors must be postulated.

The early work of Bayliss and Cannon, the recent work of Moon and of Essex and his collaborators, and our own exchange-transfusion experiments, suggest that a toxic substance or substances released from injured tissue may play a rôle in the production of traumatic shock. Various attempts to identify these substances have been unsuccessful.

The rôle of ascorbic acid in the inactivation of sympathomimetic amines. KARL H. BEYER (introduced by Walter J. Meek). *Department of Physiology, University of Wisconsin Medical School, Madison.*

Recent work on the metabolization of sympathomimetic amines has shown some of them to be excreted as such by the kidneys. Others of these compounds we have found to be inactivated both in the body and by enzyme systems. However, some of these compounds not inactivated in the presence of amine oxidase or phenol oxidase are not totally excreted as such or conjugated. From our studies of other possible modes of detoxication we have found vitamin C capable of causing inactivation of certain of these compounds both *in vitro* and *in vivo*.

In vitro, bubbling air through a phosphate buffered solution of ascorbic acid and certain sympathomimetic amines at 37°C. (pH7.0) caused the production of color and the liberation of ammonia which could be distilled from basic solution into HCl and estimated by titrating the excess HCl with NaOH. By this method we found *a*, β-phenylisopropylamine (amphetamine); *b*, β-phenyl-β-hydroxyisopropanolamine (propadrine); *c*, β-phenylpropylamine, and *d*, γ-phenylpropylamine to be deaminated with the recovery of up to 55 per cent of the ammonia, depending on the amine and the relative proportion of ascorbic acid and amine. Under similar conditions a color was produced by deamination of *e*, β (4-hydroxyphenyl) isopropylamine (paredrine) and *f*, β (4-hydroxyphenyl)-ethylamine (tyramine) did not occur to any appreciable extent.

To determine the significance of these reactions *in vivo* we studied the excretion of amphetamine as influenced by the administration of ascorbic acid. Ten milligrams of amphetamine were injected subcutaneously daily into 5 dogs. After the normal daily excretion of amphetamine was estimated for a week, 200 mgm. or, in some dogs 400 mgm., of ascorbic acid were also injected subcutaneously daily. In every case within 3 or 4 days after ascorbic acid was started the output of amphetamine began to decrease, reaching a value of 35 per cent (av.) of the controls in about 7 days. When the injections of the vitamin were discontinued the excretion of amphetamine returned within a week to the control values for the individual dog.

These results clearly suggest that ascorbic acid may play a rôle in the inactivation of certain sympathomimetic amines in the body, probably by deamination.

Acute renal hypertension produced by an amino acid. R. J. BING and M. B. ZUCKER (introduced by M. I. Gregersen). *Department of Physiology, College of Physicians and Surgeons, Columbia University, New York City.*

The isolated kidney of the cat perfused with reduced blood flow converts the amino acid l-dopa (l-dihydroxyphenylalanine) into its corresponding

pressor amine, hydroxytyramine (R. J. Bing). The following studies were undertaken to determine whether a similar reaction occurs in the completely and partially ischemic kidney in vivo. Cats anesthetized with nembutal (39 mgm. per kgm. body weight intraperitoneally) were used. The release of hydroxytyramine from the ischemic kidney was detected by its effect on the blood pressure.

In 15 experiments circulation through the vessels of both renal pedicles was completely excluded by seraphines. Collateral circulation through the renal capsule remained unimpaired. Immediately after the vessels had been clamped, 10 mgm. of dopa dissolved in 3 cc. of Ringer's solution were injected into the substance of one kidney. After 2 hours the clamp on the pedicle of the uninjected kidney was removed. No rise in blood pressure occurred. Fifteen minutes later the circulation through the injected kidney was restored. This was followed by a steep rise in blood pressure averaging 100 mm. Hg, the effect lasting for 15 minutes. Cocaine hydrochloride (6 mgm. per kgm. body weight) enhanced both the height and the duration of the response.

Seventeen experiments were performed on the partially ischemic kidney. In 8 of these 10 mgm. of dopa were injected into the renal artery; in the other 9 the injection was made directly into the renal substance. Partial reduction of the blood flow was accomplished by placing a Goldblatt clamp on the renal artery. In both series of experiments the blood pressure increased within 3 to 5 minutes after the injection. The height and duration of the rise varied with the degree of ischemia produced by the clamp. Rises amounting to 120 mm. Hg were recorded, the effect lasting from 10 to 16 minutes in different cases. Control experiments in which Ringer's solution was injected produced no elevation of the blood pressure. Likewise, there was no pressore response when dopa was injected into kidneys with normal blood flow. It is apparent that both renal ischemia and dopa are necessary for the formation of a pressor substance, presumably hydroxytyramine.

Calibration studies of the regulation of body temperature in normal dogs.[1]

J. Reese Blair (by invitation) and A. D. Keller. *Department of Physiology and Pharmacology, University of Alabama, University.*

The regulation of body temperature in twelve normal dogs was studied with a twofold purpose in mind: 1, to calibrate each animal for subsequent quantitative studies of the effect of brainstem lesions upon temperature regulation; 2, to obtain normal observations for use as a basis in determining the degree of impairment of temperature regulation in previously uncalibrated dogs possessing similar lesions.

All dogs used were adult females, varying in size from 4 kgm. to 7 kgm. body weight. Six were of the short-haired variety and six of the long-haired variety. The calibration studies were carried out by observing the rectal temperature, shivering, and respiration of each dog maintained in various thermal environments for *eight-hour* periods. Environmental temperatures used were a refrigeration box at 10°C., and heat boxes at 33°C., 36°C., and 39°C. The dogs were not fed for a period of twenty-four hours preceding any experiment so as to insure that they were in a post-absorptive state. Precautions were taken to provide proper ventila-

[1] Aided by a grant from the Rockefeller Foundation.

tion and humidity control in each case. Experimental observations were made over a period of twelve months, studies being carried out before and after the dogs had been exposed to the intense heat of the summer months.

Some correlation was observed between the pattern of the temperature regulation curve and the animal's coat of fur. Although the curves for individual dogs at any particular environmental temperature remained fairly constant, a somewhat different set of curves were found to prevail for short-haired dogs as compared to long-haired ones. The most marked differences in the two were: 1, long-haired dogs were able to maintain a more constant body temperature in the different environments employed, and 2, short-haired dogs required a greater rise in rectal temperature to initiate panting.

The use of the results in determining the impairment of temperature regulation in several dogs possessing brainstem lesions will be demonstrated.

The rôle of androgen in the production of medullary bone in pigeons by the administration of sex hormones. MARGARET A. BLOOM (by invitation), WILLIAM BLOOM (by invitation) and FRANKLIN C. MCLEAN. *Departments of Anatomy and Physiology, University of Chicago, Chicago, Ill.* (Read by title.)

As previously reported (Bloom, Bloom, Domm and McLean, 1940) we have been able to confirm the results of others by producing new bone in the medullary cavities of the long bones of pigeons, by the administration of large doses of α-estradiol, but only when the estrogen was given in the spring. Similar doses of estrogen given to male pigeons in December, or to castrate males either in December or in the spring, produced no such effect or a minimal activity in the endosteum.

When testosterone propionate was administered alone to castrate male pigeons it led to no formation of medullary bone. But when α-estradiol was administered together with testosterone propionate, the latter in doses of 0.5 to 2.0 mgm. daily, extensive medullary bone formation occurred in both male and castrate pigeons in December. Experiments are now under way to determine whether the necessary dosage of estrogen may be reduced when administered with androgen.

Hypercalcemia was produced in male and castrate pigeons, both in the spring and in December, by administration of large doses of α-estradiol. Testosterone did not enhance this effect.

Excretion and storage of bromide ion. OSCAR BODANSKY and WALTER MODELL (introduced by McKeen Cattell). *Department of Pharmacology, Cornell University Medical College, New York City.*

The excretion of bromide ion was studied *a*, in fasting dogs following single intravenous injections of sodium bromide and of sodium chloride; *b*, in dogs receiving a daily constant amount of sodium chloride and sodium bromide in their food.

Whether bromide was excreted in more than negligible amounts depended not upon the concentration of serum bromide, but upon the concentration of total serum halide (i.e., chloride plus bromide). In general, excretion did not occur at levels below 108 mM of total halide per liter of serum, in the absence of salyrgan or theophylline. The amount of bromide excreted following intravenous injection was related to *a*, the state of halide

saturation of the animal; *b*, the bromide:halide ratio in the serum, and *c*, a parameter, K, representing the ratio of the mols per cent of bromide in the urinary halide to that of bromide in the serum halide. K had a value ranging from about 0.7 to 1.0 during the period of diuresis immediately following large injections of sodium chloride or of sodium bromide; in subsequent periods, K decreased to a value of about 0.4. Salyrgan and theophylline led to considerable increases in the excretion of both bromide and chloride; K, in these instances, ranged from about 0.7 to 1.0. Sodium sulfate and large amounts of water did not appreciably increase the excretion of bromide or of chloride. In accumulation experiments in which a daily constant amount of bromide and chloride was given in food, K rose during the period of bromide storage. During the period of increasing concentration of bromide in serum storage proceeded in accordance with the distribution of this ion in the extracellular fluid.

Some properties of protyrosinase. JOSEPH HALL BODINE and THOMAS HUNTER ALLEN (by invitation). *Department of Zoology, State University of Iowa, Iowa City.*

Certain preparations catalyze the oxidation of tyramine to melanin, because they contain active tyrosinases. Other extracts do not catalyze this oxidation unless activators react with their protyrosinases. Extracts from various sources contain the enzyme, the proenzyme, or mixtures of the two.

a. A mushroom and a potato furnish tyrosinase but no protyrosinase.
b. Mealworms and grasshopper eggs yield protyrosinase but no tyrosinase.
c. Crayfish serum has protyrosinase and tyrosinase.

The protyrosinases from grasshopper egg and mealworm preparations do and do not share certain properties.

Those which they share are:

a. The power of catalysis which seems to be associated with a particular form of copper.
b. The property of being activated by egg and mealworm oils, sodium oleate, and chloroform, acetone, urethane, and urea.

Those which they do not share are:

a. The form of the activator (sodium oleate) concentration functions.
b. The ability of being activated by certain heat treatments.
c. The power of being activated by removal of electrolytes.
d. The solubilities in various concentrations of ammonium sulfate.

Thus it seems that protyrosinases are in a class of compounds whose members vary according to the source of preparation.

Impaired mobilization of liver glycogen in the absence of the anterior pituitary as a cause of insulin sensitivity.[1] R. C. DE BODO and H. I. BLOCH (by invitation). *Department of Pharmacology, New York University College of Medicine, New York City.* (Read by title.)

1. We have found a close parallelism between insulin sensitivity and resistance to the hyperglycemic action of adrenaline in partially and completely hypophysectomized dogs. Insulin, 0.025 U/kilo injected intravenously, produces hypoglycemic shock in completely hypophysectomized

[1] Aided by a grant from the American Philosophical Society.

dogs, but only a slight or negligible drop in blood sugar in normals. Adrenaline, 0.0035 mgm./kilo/min., infused intravenously for five minutes, produces marked hyperglycemia in normal dogs, but only a slight and transient effect in hypophysectomized dogs (see abstract by de Bodo, Sweet and Bloch). Partially hypophysectomized dogs are more sensitive to insulin and more resistant to adrenaline than normals, but less so than completely hypophysectomized dogs.

2. Despite this close parallelism adrenaline resistance cannot be the cause of insulin sensitivity. Adrenal-inactivated (right adrenal removed, left denervated and demedullated) and cortine-treated-adrenalectomized dogs, although showing somewhat greater response to insulin than normals, are by no means as sensitive to it as hypophysectomized dogs.

3. Both insulin sensitivity and adrenaline resistance are attributed to a common cause: impaired mobilization of liver glycogen.

A. Normal animals performing moderate exercise maintain their blood sugar level and muscle glycogen content while utilizing large amounts of carbohydrate. The sugar utilized in the muscles is drawn from the blood sugar which is replenished from liver glycogen. Since this liver glycogen mobilization occurs in the absence of adrenal medulla and liver nerves, factors other than these two must be involved (This Journal **123:** 18, 1938).

B. Recovery from insulin hypoglycemia occurs at the expense of liver glycogen. Here too adrenaline and liver nerves are not essential to mobilization of liver glycogen; again other factors must be involved.

C. In hypophysectomized animals given minute amounts of insulin there is an abrupt fall of blood sugar to hypoglycemic levels without recovery. This reveals that liver glycogen (present in large quantities) is not mobilized by the factors normally effective as in A and B. Adrenaline liberated at a certain level of hypoglycemia is also ineffective in mobilizing liver glycogen.

Conclusions: the factors normally effective in mobilizing liver glycogen are ineffective in the absence of the anterior pituitary, and this is an important cause of the sensitivity to insulin observed in hypophysectomized animals.

The rôle of the anterior pituitary in adrenaline hyperglycemia and liver glycogenolysis.[1] R. C. DE BODO, J. E. SWEET and H. I. BLOCH (by invitation). *Department of Pharmacology, New York University College of Medicine and Department of Surgical Research, Cornell University Medical College, New York City.* (Read by title.)

When adrenaline, 0.0035 mgm./kilo/min., is infused intravenously for five minutes into normal dogs in the post-absorptive state a prompt and marked rise in blood sugar occurs. Within fifteen minutes from the beginning of the infusion the blood sugar reaches its maximum, an increase of 70–90 per cent, and then gradually returns to normal within 60–120 minutes. In contrast to this, hypophysectomized dogs under identical conditions show only a slight rise in blood sugar, the maximum increase being only 12–24 per cent and the blood sugar returning to normal within 30–45 minutes.

The absence of a marked hyperglycemic response in the hypophysectomized dogs can not be attributed to a lack of liver glycogen, since several

[1] Aided by a grant from the American Philosophical Society.

of these animals were sacrificed and their liver glycogen content was found to be 3.8–4.5 per cent, well within normal limits. Furthermore some of the animals died in spontaneous hypoglycemic shock and had 1.2–2.0 per cent liver glycogen content a few hours after their death. Some died in experimentally induced insulin shock and had 2.7–2.9 per cent liver glycogen. On the other hand, normal dogs fasted for 7–14 days, having smaller amounts of liver glycogen than the hypophysectomized animals, showed a greater response to adrenaline. Normal dogs fasted for such a length of time were found to contain 1.0–1.3 per cent liver glycogen and yet showed an increase of 30–60 per cent in blood sugar when adrenaline, 0.0035 mgm./kilo/min., was infused intravenously for five minutes.

In another series of dogs in which only the neurohypophysis had been destroyed and the anterior pituitary left intact, producing permanent diabetes insipidus, adrenaline infused intravenously in the same quantities produced the same effect as in normal animals.

Since there is hardly any increase in blood sugar in the hypophysectomized animal in response to an amount of adrenaline given intravenously which produces a marked hyperglycemic reaction in normal dogs and since this absence of response in the hypophysectomized dog is not due to a lack of liver glycogen, it is concluded that in the absence of the anterior pituitary the liver glycogen is not readily mobilized by adrenaline.

The influence of the thyro-parathyroid glands on a remaining kidney.[1]

E. L. Borkon (introduced by M. A. Hinrichs). *Department of Physiology and Health Education, Southern Illinois Normal University, Carbondale, Ill.*

Thirty-day old albino rats were subjected to a right nephrectomy and one half of these had the thyroid and the parathyroid glands removed using the method described by Templeton (Endocrinology **21**: 541, 1937). Immediately after operation and for the next one hundred days both groups were fed a diet of Purina Fox Chow, ad libitum. On the hundredth postoperative day, all the rats were sacrificed and the remaining left kidney of each weighed, after being stripped of its capsule. The left kidneys were then either burned to obtain total ash weights or were prepared for histological examination. Wet weights of the left kidneys in the first group of experiments were as follows:

	AVERAGE KIDNEY WEIGHT	RANGE OF KIDNEY WEIGHTS	AVERAGE BODY WEIGHT	RANGE OF RAT WEIGHTS	KIDNEY PER KILOGRAM BODY WEIGHT
	grams	*grams*	*grams*	*grams*	*grams*
Right nephrectomy and thyroparathyroidectomy	0.917	0.855–0.972	211	190–243	4.346
Right nephrectomy only	1.595	1.321–2.012	275	250–320	5.799

Histological and ash findings are not yet complete. Other experiments are already begun in which one kidney was removed and the effects of feeding 0.2 per cent desiccated thyroid upon the remaining kidney were studied. Still other experiments in progress repeat the above experiments but leave both kidneys intact. At this time 59 rats have been observed.

[1] Aided by a grant from the Ella Sachs Plotz Foundation.

Correlation of potency of urine extract to inhibit gastric motility with potency to inhibit secretion. J. E. BOURQUE, JR. (by invitation), M. H. F. FRIEDMAN (by invitation) and T. L. PATTERSON. *Department of Physiology, Wayne University College of Medicine, Detroit, Mich.*

These experiments were undertaken to determine whether the inhibition of gastric secretion and inhibition of gastric motility by urine extracts were due to the same active principle. Relatively non-toxic and pyrogen-free extracts prepared from urine of normal individuals and of patients with gastro-intestinal pathologies, were employed. These extracts were administered intravenously to gastric fistula dogs previously fasted for 48 hours and the contractions recorded by the balloon-bromoform manometer method. Control experiments consisted of recording gastric motility without administration of the extracts, as well as with injections of a placebo. The effect of the extracts on gastric secretion was studied in dogs with a Heidenhain pouch.

The latent period for the inhibition of gastric motility was found to be approximately the same for all extracts of normal male urine prepared by the same procedure. However, the duration of the inhibition exerted by the extracts varied with the preparation. The latent period for the inhibition of gastric motility was shorter than for the inhibition of gastric secretion. The degree and duration of the inhibition of gastric motility appear to be correlated with the degree of inhibition of gastric secretion by the same extract. However, after continued injections of urine extracts, two dogs became refractory to previously adequate doses. This prevents any emphasis of the quantitative aspects of the results. Recheck experiments with freshly prepared dogs, indicate that the data obtained previous to the onset of the refractory state are probably correct.

Variations in diastolic volume and stroke output of the ventricles with the phases of respiration. T. E. BOYD and MARY C. PATRAS (by invitation). *Department of Physiology and Pharmacology, Loyola University School of Medicine, Chicago.*

A cardiometer was used, the chest being closed and natural breathing allowed to go on. The recording device was a sensitive tambour, its membrane inclosed on both sides. The air chamber on one side is connected to the cardiometer, that on the other to the intrapleural space of the animal. Respiratory excursions of the membrane are too small to interfere seriously with cardiometric recording, and external pressure on the ventricles follows the normal respiratory changes of intrathoracic pressure.

With inspiration, diastolic volume increases. Combined stroke output also increases, reaching a maximum at the end of inspiration or with the first systole following the onset of expiration. With expiration diastolic volume and stroke output are reduced gradually, reaching a steady level if the expiratory pause is sufficiently prolonged. These effects are exaggerated by the deep and prolonged inspiration of the vagotomized animal, also by obstruction of the trachea.

Earlier workers have reported a reduction of stroke output during inspiration, but the procedure followed left the ventricles under constant atmospheric pressure. We obtain the same result under similar conditions. It evidently is due to abnormal resistance to ventricular filling,

especially during inspiration, blood being dammed back into the auricles and veins. If the recording tambour is shut off from connection with the thoracic cavity, and opened to outside air, venous pressure rises and its respiratory fluctuations are reduced.

Action potentials of visceral smooth muscle. EMIL BOZLER. *Department of Physiology, The Ohio State University, Columbus.*

Monophasic action potentials of various types of visceral smooth muscle were studied by means of a direct coupled amplifier. The discharge of this type of muscle normally consists of brief impulses, but the peristaltic waves of the ureter are accompanied, in most species, by a negative variation which is sustained for some time as in cardiac muscle. In the ureter of the guinea pig there is, instead, a discharge of high frequency, but this discharge usually is superimposed on a slow negative variation which may persist after all signs of rhythmic activity have disappeared and is accompanied by continued activity of the muscle. Such action potentials may be considered as intermediate between a repetitive discharge of discrete spike potentials and the potential change with a plateau in which the impulses have fused entirely. A retention of negativity following a spike potential also may appear in the discharge of uterine muscle. A positive after-potential was found under certain conditions. Slow potential changes which run parallel with the mechanical response such as are often described in the literature were not observed in experiments with isolated muscle preparations.

Effect of angiotonin on circulatory dynamics.[1] STANLEY E. BRADLEY and BARBARA ANN PARKER (introduced by H. W. Smith). *Departments of Physiology and Medicine, New York University College of Medicine, New York City.* (Read by title.)

A purified preparation of angiotonin, the pressor principle which Page and his collaborators have suggested as the chemical mediator in both experimental and clinical hypertension, was supplied to us through the kindness of Drs. Page, Corcoran and Helmer, and in addition to other studies we thought it of interest to examine its effects on the cardiovascular system of normal subjects.

Cardiac output was measured by the ballistocardiograph of Starr *et al.*, and simultaneous records of the radial arterial pressure were recorded with a Hamilton membrane manometer. The electrocardiogram was recorded with a string galvanometer. Mean arterial pressure was calculated from the area under the blood pressure tracing as measured with a planimeter, and mean effective peripheral resistance was calculated in c.g.s. units from Frank's classical equation. The angiotonin was given intravenously in one or two cc. doses.

In no instance was there any subjective response. Ten to 15 seconds after injection the blood pressure rose sharply, systolic pressure rising much more than diastolic. Cardiac output was markedly reduced and did not recover to its control value until the pressor effect had worn off. The reduction in cardiac output was largely the result of slowing of the heart (? vagal inhibition), the stroke volume falling slightly. The intravenous injection had no immediate effect on the ballistocardiographic record or

[1] Aided by a grant from the Commonwealth Fund.

the electrocardiogram. Peripheral resistance rose sharply to more than double its control level. All changes were qualitatively alike, but more marked and of longer duration following the larger dose.

The fact that the pulse pressure increases markedly despite a slight decrease in stroke volume suggests that angiotonin decreases the volume elasticity coefficient of the arterial reservoir. This question is being subjected to further examination.

Changes in pH and the rate of flow of saliva accompanying pH changes in arterial blood during acetyl choline stimulation of the submaxillary gland.[1] CHARLES R. BRASSFIELD and A. P. HANDS (by invitation). *Department of Physiology, University of Michigan, Ann Arbor.*

Studies were made of changes in pH and rate of flow of saliva from the submaxillary gland of the dog in response to changes in pH of arterial blood induced by changing O_2 and CO_2 tensions and by injecting acids and bases. Salivary secretion was produced by continuous intra-arterial injection of acetyl choline and pH changes were followed continuously in both blood and saliva by means of glass electrodes. Rate of saliva flow was followed with a Gibbs drop recorder.

Lowering the O_2 tension of the inspired air produced parallel increases in saliva and blood pH but the saliva change was less. Saliva flow was increased at the beginning of the administration but diminished toward the close of the procedure. Increasing the CO_2 tension of the administered gas produced pH decreases in both blood and saliva with the change in saliva being less. Saliva flow was decreased. Overventilation produces an increase in flow of saliva and an increase in its pH which is less than the change produced in blood. Lactic acid injections decreased the flow of saliva and its pH; sodium bicarbonate increased the flow but decreased the pH.

It is noteworthy that an increase in saliva flow was accompanied or preceded by an increase in the difference between the saliva pH and arterial blood pH. Those procedures which caused a greater blood pH increase than in saliva increased the flow while those procedures which caused a greater blood pH decrease than in saliva decreased the flow. This suggests that the relation of intracellular to extracellular hydrogen ion concentration plays an important rôle in the response of the gland to acetyl choline stimulation. Tentative analysis indicates a possible application of this principle to the activity of the respiratory neurones in the control of breathing.

Chemical initiation of rhythmic local responses in nerve preceding trains of propagated impulses. FRANK BRINK, JR. (by invitation) and D. W. BRONK. *Department of Physiology and Biophysics, Cornell University Medical College, New York City.*

Our previous studies of chemical excitation of nerve have related the average frequency of conducted impulses in single fibers to the degree of chemical change. To analyze further the mechanism of this excitation we have recorded in a single fiber the electrical changes occurring in the chemically altered region of the nerve. Thus far technical difficulties have limited this type of experimental approach to the giant axon of the Squid.

[1] Aided by a grant from the Rackham Foundation.

One recording electrode was in contact with the giant axon near one end. The other made contact through a pool of sea water covering about 3 mm. of the axon at a point well removed from either end. The chemical excitation to be described was produced by removing the calcium and magnesium from this sea water.

The changes in structure of the nerve thus produced give rise to fluctuating circulating currents in the chemically treated region. They are recorded as a cyclic change of potential difference between the recording electrodes. It is characteristic of these chemically induced local potentials that they are definitely rhythmic and that they periodically wax and wane in amplitude. Their frequency is of the order of 300 per second. Their amplitude is larger the greater the degree of change in the chemical environment. When under appropriate conditions the amplitude attains a certain critical value a train of conducted impulses develops from the negative peaks of the local potential cycles. The initial frequency of the conducted impulses is the same as the final frequency of the preceding local responses. Each of the rhythmic local circulating currents apparently acts as the stimulus which initiates each of the successive conducted impulses.

Arvonataki has shown that such local oscillatory potential changes occur when a nerve with reduced calcium content is cathodally polarized by a direct current. Our experiments demonstrate these rhythmic local changes under the mere removal of calcium. Further investigation is required to determine whether a small direct current arising from an intrinsic source of emf. participates in this process of chemical excitation.

The effects of sterol feeding on arterial blood pressure in rats. H. L. Briskin (by invitation, R. F. Stokes (by invitation) and C. I. Reed. *University of Illinois, College of Medicine, Chicago, Ill.*

An extensive series of sterols is being studied but only those experiments with 7-dehydrocholesterol are sufficiently complete for any conclusions. Blood pressure was measured at frequent intervals in the tail. Doses of sufficient size to cause loss of weight did not induce any changes in blood pressure except an insignificant tendency to depression. The sterol was fed at several ranges from 10,000 International Units of Vitamin D daily to 100,000 units.

Effects of autonomic stimulation on the estrous cycle of the rat. L. P. Britt (introduced by S. W. Britton). *Physiological Laboratory, University of Virginia Medical School, University.* (Read by title.)

One hundred animals were subjected to psychic stimulation (motor horn or air jet sound, five minutes twice daily), adrenalin injection (2.5 cc. 1:100,000, twice daily), or served as controls. Vaginal smears were observed daily (10 to 20 days), and only those which contained cornified cells were considered as positively indicative of estrus. The estrous cycles of all experimental animals were studied, moreover, during pre-experimental periods of 10 to 20 days.

The average percentage of days spent in estrus was found to be 39.7 in control animals, while in rats subjected to psychic stimulation the percentage rose to an average of 47.2, and was attributable to a shortening of the cycle. An even more marked increase in the percentage of the total

experimental period spent in estrus was observed in those rats which were treated with adrenalin. In this case the average percentage of days in estrus was 62.2. However, this increased estrual activity was found to be due to a prolongation of the individual cycles together with an increased length of the estrual phase.

Rats from which the cervical and lumbo-abdominal sympathetic chains were excised showed, on the other hand, a decrease in the average number of days spent in estrus (3.4 as compared to 7.9 for the controls), as well as a decrease in the percentage of days in estrus. Moreover, psychic stimulation and adrenalin failed to evoke any estrogenic response in such sympathectomized rats.

It was concluded that psychic stimulation or adrenalin injection may significantly alter the normal continuity of the estrous cycle of the rat. The effect of such autonomic stimulation may thus be said to be estrogenic in character. On the other hand, cervico-abdominal sympathectomy served to prevent such action.

The influence of hypothalamic lesions on pancreatic diabetes. J. R. BROBECK (by invitation) and C. N. H. LONG. *Department of Physiological Chemistry, Yale University School of Medicine, New Haven, Conn.*

Davis, Cleveland and Ingram (Arch. Neurol. and Psychiat. **33**: 592, 1935) have reported that bilateral hypothalamic lesions in cats, particularly those involving the region around the fornix, are followed by an atypical diabetes when the pancreas was subsequently removed.

In a series of six partially depancreatized rats with a daily glycosuria of between 5–9 grams bilateral lesions were placed in the hypothalamus with a Horsley-Clarke stereotactic instrument. In none of the animals was any alleviation of the degree of diabetes, as judged by the daily glycosuria, observed. The food intake was kept constant throughout.

The lesions were placed bilaterally in the regions of the fornices, at levels varying from the anterior border of the chiasma to the level of the median eminence. Certain of these lesions destroyed areas comparable to those described by Davis, Cleveland and Ingram in their cats. In one animal both paraventricular nuclei were severely damaged.

In the above series of animals the lesions did not result in an increased body weight. In another partially depancreatized rat that was not exhibiting glycosuria, the placing of bilateral lesions at the base of the hypothalamus between the levels of the posterior border of the chiasma and the median eminence, 1 mm. on each side of the midline, was followed by a greatly increased appetite, rapid gain in weight and the appearance of glycosuria. After a period of 25 days the food was reduced to the preoperative (hypothalamic) level, but the glycosuria continued although at a lower level. On exposure of the animal to successive periods of high food intake and rapid gain in weight, the severity of the diabetes as judged by the degree of glycosuria on the basal diet steadily increased. However, when the rat reached a weight of over 500 grams the glycosuria spontaneously disappeared.

Chemical control of respiration and activity in peripheral nerve. D. W. BRONK, FRANK BRINK, JR. (by invitation) and P. W. DAVIES (by invitation). *Department of Physiology and Biophysics, Cornell University Medical College, New York City.*

These experiments are concerned with the influence of various chemical agents on the oxygen consumption of nerve and its relation to the degree of activity in the nerve.

Isolated frog nerves were placed in a chamber in which the oxygen consumption was measured with the method described by Davies and Brink in these communications, and the action potentials were recorded at the same time. The entire nerve was immersed in solutions of various chemical compositions.

Lowering the concentration of calcium ions to one millimolar has produced no measurable change in oxygen consumption and no detectable impulses. Reduction of the concentration to 0.5 millimolar causes a 20 to 40 per cent increase in oxygen consumption although no propagated nerve impulses have been recorded. It is not impossible, however, that unobserved impulses were initiated in small C fibers. With further decreases in the concentration of the calcium ion impulses were observed in increasing numbers and there was a further augmentation of the rate of oxygen consumption to more than twice the resting value. If, now, sodium azide or chloretone in appropriate concentrations were added to the solution, the oxygen consumption could be restored to the resting level as the impulse discharge disappeared.

There are some instances however in which suppression of activity by an appropriate chemical agent does not reduce the rate of respiration. For instance, 20 millimolar potassium chloride abolishes all evidence of the impulse discharge initiated by the previous removal of calcium and at the same time actually causes a further increase in the rate of oxygen consumption.

These experiments show that oxygen consumption and the initiation and propagation of impulses in nerve can vary independently.

Reduction of sexual behavior in male guinea pigs by hypothalamic lesions.

J. M. Brookhart and F. L. Dey (introduced by S. W. Ranson). *Institute of Neurology, Northwestern University Medical School, Chicago, Ill.*

The influence of hypothalamic lesions upon the sexual behavior of male guinea pigs was studied in nine adult animals. The normal pattern was established preoperatively in five animals by observing their behavior when placed singly with one or more estrous females. Electrolytic lesions were placed in the hypothalami between the optic chiasma and the infundibulum. Sexual behavior with estrous females was observed in all nine animals postoperatively. The libido and fertility of the males was also tested by placing each male with three normal females for a sufficient length of time to allow each of the females to go through two estrous cycles. The brains, seminal vesicles and testes were prepared for histological examination.

Sexual behavior was abolished postoperatively in four animals; in three animals sexual activity was greatly reduced; and in two animals the behavior pattern was normal. The seven animals which showed reduction of sexual activity produced three pregnancies out of the total of forty estrous periods which occurred in the twenty-one females. The two animals which showed normal behavior produced four pregnancies out of the total of ten estrous periods in their six females. The results of electrical ejaculation tests and epididymal smears, as well as the histological condition of the testes and the seminal vesicles indicated no impairment of endocrine

activity on the part of the testes or the anterior pituitary. The lesions in the two cases which showed normal behavior were significantly different from those in the animals which showed reduced sexual activity. It is suggested that the reduced sexual activity was the result of the destructive lesion in the hypothalamus.

A locally constructed Dale-Schuster double perfusion pump with modifications in construction. BRUCE T. BROOKMAN (by invitation) and H. MORROW SWEENEY. *School of Medical Sciences, University of South Dakota, Vermillion.* (Demonstration.)

This pump represents our attempt to construct the Dale-Schuster double perfusion pump, and incorporates certain changes in construction which lead to simplification of the construction and to a more readily controllable output.

A perfusion range is shown by the pump for blood of from 0 to 300 cc. per minute, at 150 mm. Hg, and a correspondingly greater capacity at the lower perfusion pressures. Due to a change in the diaphragm construction, calculation of flow volume at any fulcrum setting, with any peripheral resistance, without the use of a stromuhr in the perfusion circuit is more readily made.

The effect of various brain lesions on morphine-induced hyperglycemia and excitement in the cat. CHANDLER MCC. BROOKS, ROBERT A. GOODWIN[1] (by invitation) and HAROLD N. WILLARD (by invitation). *Department of Physiology, the Johns Hopkins University, School of Medicine, Baltimore, Md.*

Bodo, CoTui and Benaglia (1936, 1937) found that the marked rise in blood sugar which occurs following administration of morphine is due primarily to generalized discharge of the thoraco-lumbar division of the autonomic system and the release of adrenin. In cats morphine also produces excitement and increased somatic activity. In chronic spinal cats (Bodo and Brooks, 1937) morphine fails to produce a rise in blood sugar; furthermore, those portions of the body caudal to the level of cord section give no indication of cord excitation. The following experiments were undertaken in attempting to localize more exactly the site of the excitant action of this drug.

Three chronic preparations from which the neocortex of both hemispheres had been removed showed typical somatic signs of excitement and rises in blood sugar comparable to those obtained before operation.

In eight cats which had been decerebrated at the mid-collicular level twelve to twenty-four hours before administration of morphine no rise in blood sugar was obtained. Some activity and variations in intensity of the decerebrate rigidity were observed but nothing occurred which could be classified definitely as morphine-induced excitement.

Lesions were placed in the preoptic areas and in the anterior portion of the hypothalamus in seven cats by means of a Horsley-Clarke stereotaxic instrument. These animals were injected with morphine four days, two weeks, and in two cases, one month after operation. Four individuals, including the two cats which were kept for one month, showed a morphine-induced hyperglycemia which was somewhat less than that observed before

[1] Henry Strong Denison Scholar.

operation but in the remaining four the rise was comparable. Those lesions though modifying certain responses did not prevent morphine excitement.

Massive injury to the posterior portions of the hypothalamus practically abolished the morphine effect in blood sugar but somatic excitement occurred. This evidence indicates that morphine produces an effect on blood sugar level by stimulating autonomic centers so located that they are inactivated by lesions involving the posterior areas of the hypothalamus.

The angle method for blood sedimentation. CLYDE BROOKS. *School of Medicine, Louisiana State University, New Orleans.*

Heretofore, the blood sedimentation test has been made with the tubes placed more or less accurately in the vertical position. However, it has been found that even a slight deviation of the tube from the upright position caused a distinct increase in the sedimentation rate.

However, there are advantages in making the sedimentation test with the tubes set up at an angle. The angle method is more rapid. A slight variation from the 45° position does not cause a marked change in sedimentation rate.

Using the angle method, slightly altered standards of criteria for normal and pathological sedimentation curves have been established. Studies on the effect of certain drugs and certain diseases on sedimentation rate are summarized.

Differences in rates of O_2 or CO consumption of fertilized and unfertilized *Arbacia* eggs as influenced by methylene blue. MATILDA MOLDENHAUER BROOKS. *University of California, Berkeley, and The Marine Biological Laboratory, Woods Hole, Mass.*

These experiments were done to obtain more information on the mechanism of methylene blue action in cells. Eggs were used because they are not complicated by the presence of hemoglobin. The O_2 or CO consumption by fertilized and by unfertilized *Arbacia* eggs was measured by means of the Warburg manometer method, using either air or 100 per cent CO in the presence or absence of methylene blue in final concentrations of 0.0001 to 0.00001 M. Fertilized eggs were used ½ hour after addition of sperm. The volume in cubic centimeters of O_2 or CO consumed before and after the addition of methylene blue (m.b.) is shown in a typical series in table 1.

These results show that 1, the rate of CO consumption by fertilized and

TABLE 1

Average volumes of O_2 or CO consumed by equal numbers of Arbacia eggs

	UNFERTILIZED EGGS			FERTILIZED EGGS		
	First 95 min.		Second 95 min.	First 110 min.		Second 110 min.
	No. of expts.	Ccm.	Ccm.	No. of expts.	Ccm.	Ccm.
CO	3	0.51	0.74 (with m.b.)	6	0.55	1.15 (with m.b.)
Air	2	0.3	0.5 (with m.b.)	6	1.8	4.5 (with m.b.)
CO	3	0.53	0.57 (no m.b.)	2	0.62	0.57 (no m.b.)
Air	3	0.12	0.23 (no m.b.)	1	2.0	4.4 (no m.b.)

unfertilized eggs was the same; 2, m.b. increased the rate in both cases, and to a greater degree with fertilized eggs; 3, m.b. appears to have little effect in these concentrations upon the rate of O_2 consumption in air in both cases; 4, the relatively low rate of O_2 consumption of unfertilized eggs is confirmed; 5, CO inhibited cleavage mainly beyond the 2-cell stage and m.b. had no effect on this.

Upon the presence and distribution of a chromatophorotropic principle in the central nervous system of Limulus. F. A. BROWN, JR. and ONA CUNNINGHAM (by invitation). *Northwestern University, Evanston, Ill.* (Read by title.)

Cells and cell groups which, from their histological picture, probably have endocrine activity, are being described in the nervous system of many vertebrates and invertebrates. So far, their functional significance is almost completely unknown. The experiments to be described demonstrate the presence and distribution of a chromatophorotropic principle in the nervous system of Limulus.

The central nervous system of Limulus polyphemus was divided into portions which were extracted and tested for chromatophorotropic activities upon black and white chromatophores of Uca pugnax and isolated red and white chromatophores of Cambarus. All portions of the nervous system which were tested showed chromatophorotropic activity with the Uca black and white chromatophores and Cambarus white ones. Using the weights of the various portions of the nervous system and the relationship between the relative concentration of active principle and the magnitude of the chromatophorotropic effect, the concentration of the active principle in the various portions of the nervous system was readily calculated. It was found that *1*, the posterior portion of the circumesophageal nerve ring possessed the greatest concentration (arbitrarily called 38.5), followed in order of decreasing concentrations by *2*, the lateral portions of the nerve ring (18.6); *3*, the fifth ganglion (11.3), fourth ganglion (10.7), anterior portion of the ring (10.0), seventh ganglion (8.3), and *4*, the sixth ganglion (4.1). Evidence is submitted suggesting that the cells producing the active principle are distributed throughout the nerve ganglia rather than restricted to a single site within the nervous system. All the effects observed upon the three types of chromatophores which yielded positive results suggest that a single principle is involved in these three instances. The activity of the principle from the central nervous system of Limulus upon the chromatophore types was compared with the activities of other known arthropod hormones and it was concluded that the Limulus principle differed from all the other arthropod principles thus far known with the possible exception of a secretion of the brain of certain insects.

Preliminary studies on metabolism of tissue cultures. AUSTIN M. BRUES and HILDEGARD WILSON (by invitation). *Collis P. Huntington Memorial Hospital of Harvard University, Boston, Mass.*

Investigation of the metabolic activity of tissues in culture has certain advantages over other methods of studying tissue metabolism. In tissue culture one is dealing with a system in which cell surfaces are intact and localized intracellular reactions take place as in the living organisms. Moreover, the system is capable of prolonged life, and growth is a controllable factor. Certain aspects of the carbohydrate metabolism of chick

embryo tissues have been studied by means of analyses of media and observations on cells grown in roller bottle cultures.

Sugar utilization by cultures given 100 mgm. glucose per cent in the medium continues until it is exhausted. If the amount is increased fivefold, utilization is at first more rapid, but ceases while glucose is still present. That this is not due to accumulation of metabolites is suggested by the fact that a considerably higher total glucose utilization occurs in cultures to which glucose is added daily, while the medium is otherwise unchanged. Lactic acid is formed in both the presence and absence of glucose; its formation is increased as greater amounts of glucose are used. There is some evidence for subsequent utilization of the lactate first formed.

Added pyruvate is rapidly metabolized by these tissues but growth does not take place if pyruvate is substituted for glucose. Increased glucose utilization in a medium rich in glucose does not increase the apparent growth rate of the tissue. Insulin has no effect on glucose utilization either at high or at low levels in the medium. Colchicine in a concentration which arrests all cell division has no effect on glucose utilization.

If cultures are grown in a medium containing nitrogen in the form of amino acids, its utilization by the cells can be demonstrated.

The effect of experimentally produced obesity on energy and water metabolism.[1] JOHN M. BRUHN and A. D. KELLER. *Department of Physiology and Pharmacology, University of Alabama, University.*

The energy and water metabolism of three dogs was studied before and after diabetes insipidus and obesity was precipitated by hypothalamic puncture. The animals were fully conditioned to cage life and to a constant amount of standard diet for several weeks before operation. The same food allowance was continued during the postoperative period except for an interval of 22 weeks when the animals were kept in an outside yard. During this time the food intake was not measured but an unlimited amount was not available. The body weight of two animals had nearly doubled and had doubled in a third by the end of the yard confinement (thirty five weeks after operation), and has been maintained subsequently with the preoperative food level and cage regime. The last heat production studies were made a year after operation.

The total heat production of two of the obese animals was considerably greater than the preoperative values. This augmentation with increased weight is comparable to that of normal animals of equivalent weights and thus the heat production of these two obese animals agrees with that of normal non-obese dogs of corresponding weight. The increase of metabolism of the third obese animal, whose weight increment was greatest (doubled), was proportionately greater than that of the other dogs. It is evident therefore that under these conditions fat deposition is accompanied by a total metabolism equal to or greater than that of tissue deposited during normal growth. The question arises as to whether the rate of metabolism of the recently acquired fatty tissue is the same as that of the body tissue of a normal dog of comparable weight or whether its rate is lower but demands extra energy expenditure from the non-adipose tissue.

The total fluid exchange increased in direct proportion to the increase in body weight and not in proportion to the increase in heat production.

[1] Aided by a grant from the Rockefeller Foundation.

Accordingly the water exchange per unit of body weight has remained constant.

The influence of boric acid on the survival of excised muscle. E. H. BRUNQUIST (introduced by R. W. Whitehead). *Department of Physiology and Pharmacology, University of Colorado School of Medicine, Denver.*

In a former report (Am. J. Physiol. **119:** 282, 1937), prolonged survival of excised frog sartorius muscles was described. Survivals of over a hundred days at 5° to 6°C. and of as long as 12 days at 25° were obtained without the addition of any bacteriostatic agent to the $NaHCO_3$-buffered Ringer's fluid.

In later experiments it has been found that survival may be prolonged by the addition of boric acid to the Ringer's fluid. The optimal concentration varies with the temperature at which the muscles are maintained. Support of survival is especially conspicuous at the higher temperatures (e.g., at 25°), and in the case of muscles from the less hardier batches of frogs.

The hemoglobin method for the determination of pepsin in gastric drainage. G. R. BUCHER (by invitation) and J. M. BEAZELL. *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

The superiority of the hemoglobin method for the determination of pepsin is evident because, in contrast to other methods, it employs a stable and reproducible substrate. However, when applied to gastric drainage, the original method described by Anson and Mirsky required variable dilution of samples, since the method was designed for quantities of pepsin much smaller than those commonly encountered in conveniently measurable volumes of gastric juice. This constituted a disadvantage in two respects. First, the degree of dilution necessary to bring the sample within the range of the method could be determined only by preliminary trial; and second, "inhibitor substances" are affected to a greater extent than the enzyme; thus dilution modifies the effective activity of samples in a variable manner. A modification suggested by Beazell *et al.* partially overcame these disadvantages, but subsequent experience has shown that with this method, also, it is necessary to dilute many samples of human gastric juice in order to bring the activity within the range of the method. Therefore the procedure has been further modified so that the range will embrace all peptic potencies likely to be encountered in undiluted gastric juice. This has been accomplished by tripling the quantity of substrate, decreasing the digestion period to 7.5 minutes, and using 0.5 cc. of undiluted gastric juice. The unit as previously defined remains unchanged.

In 24 experiments on 4 vagotomized total pouch dogs, the average minute output in response to 1 mgm. of histamine was 0.81 cc. and 111 milli-units of pepsin during the first half-hour; 1.02 cc. and 62 milli-units of pepsin during the second half-hour. Experiments to date indicate that in the human, the response to 0.5 mgm. histamine is about thrice that of the dog with respect to volume output per minute while the pepsin output per minute is about 50 times as great.

In vitro removal of pyruvic acid in human blood.[1] ERNEST BUEDING and ROBERT S. GOODHART (introduced by Norman Jolliffe). *Medical Service of the Psychiatric Division, Bellevue Hospital and Department of Medicine, College of Medicine, New York University, New York City.*

Oxalated human blood incubated at room temperature for thirty minutes removes 30 to 70 μ grams of added pyruvic acid per ml. blood. This removal is increased by inorganic phosphate (pH optimum: 7.4) decreased by Cl. Mg-ions and heparin and unaffected by thiamin, cocarboxylase, citrate and the oxygen tension of the blood. Defibrinated blood removes much less pyruvic acid than oxalated blood. Blood cells remove considerably less pyruvic acid than whole blood, while plasma is completely inactive. Cells previously heated for two minutes at 100°C. are completely inactive, even when unheated plasma is added. The addition of heated plasma to unheated cells restores the activity to at least 70 per cent of that of whole blood. The removal of pyruvic acid is therefore effected by an enzyme system consisting of thermolabile components within the blood cells and of thermostabile factors in the plasma.

The increased removal of pyruvate caused by NaCN is not due to an activation of an enzyme system, but to the formation of pyruvic acid cyanhydrine. The disappearance of pyruvate is not associated with the formation of other carbonyl compounds. In the presence of NaF and under anaerobic conditions about 90 per cent of the removed pyruvate is recovered as lactate; in the absence of NaF more lactate is formed than pyruvate disappears, indicating an analogy between the glycolytic enzyme system in blood and in muscle.

Some effects of pregneninolone in the experimental animal. E. S. BURGE (introduced by A. C. Ivy). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

The effects of progesterone administered subcutaneously were compared with those of pregneninolone given orally and subcutaneously. Uterine strips from immature rabbits treated with sensitizing doses of estrone and then progesterone did not respond to 0.01 to 10 units of pituitrin. Strips from estrone-oral pregneninolone-treated animals showed a marked increase in tone and magnitude of contractions—similar to the effect obtained with estrogens.

Castrated female rats of a strain having vestigial ventral prostate glands were given estrone and then 3–20 mgm. of pregneninolone orally and 1–5 mgm. subcutaneously. Histologic sections of the vaginae showed changes from mucification to early cornification, indicative of estrogen effect. Yet, in sections of the ventral prostates of these same animals "light areas" indicative of androgenic activity were found.

Electrical theory of sleep, consciousness, and unconsciousness. W. E. BURGE and E. L. BURGE (by invitation). *Department of Physiology, University of Illinois, Urbana, and 93rd Bombardment Squadron, March Field, Calif.*

When one nonpolarizable electrode was placed on the scalp of the forehead and another on the forearm with a galvanometer in the circuit, a current of low amperage passed from the scalp to the forearm thus showing the scalp to be positive. During sleep this current decreased practically

[1] Aided by The John and Mary R. Markle Foundation.

to zero. When the subjects woke and became active, it increased, in some instances to 0.925 microamperes.

It has been shown (Burge et al. 1910–40) that during anesthesia, more negative charges leave the brain by efferent nerves than pass to it by afferent nerves, resulting in a loss of negative charges, thereby rendering the brain cortex positive and hence less irritable (anelectrotonus) with resulting unconsciousness and anesthesia. During recovery from anesthesia more negative charges pass to the brain by afferent nerves than leave by efferent nerves resulting in a gain of negative charges thereby rendering the brain cortex electronegative and hence more irritable (catelectrotonus) with resulting recovery from anesthesia and regaining of consciousness. Forbes (1922) found that ether anesthesia decreased the number of negative charges passing to the brain over the sensory roots of the spinal nerves by blocking them at the first synapse in the cord.

It has also been shown that the scalp of the conscious dog, like that of the human, is electropositive, and that during anesthesia this positive potential diminishes with the decrease produced in the negative potential of the underlying cerebral cortex, and during recovery from anesthesia the positive potential of the scalp rises with the increase in the negative potential of the underlying cortex, hence scalp potential is an index to cortical potential.

The decrease observed in the positive potential of the human scalp during sleep indicates a fall in negative potential and hence in irritability of the underlying brain cortex with resulting unconsciousness and sleep, similar to what occurs during anesthesia. The rise in the positive potential of the scalp upon waking indicates an increase in the negative potential and hence in irritability of the underlying cortex, with resulting waking and regaining of consciousness, similar to what occurs during recovery from anesthesia.

Effects of exercise, rest and sleep on scalp potential. W. E. Burge and M. J. Vaught (by invitation). *Department of Physiology and School of Physical Education, University of Illinois, Urbana.* (Read by title.)

When one nonpolarizable electrode was placed on the scalp of the forehead and another on the forearm a current of low amperage passed from the scalp to the forearm. This current increased with physical activity and decreased with rest.

The current was weakest during sleep, around 0.01 microamperes. When the subject awoke it increased to around 0.05 microamperes. Upon getting up the current increased still further, how much depended on how active the subject became.

Stoking the furnace and removing the ashes increased the current from the low resting level of 0.07 microampere to 0.15 microampere. Climbing four flights of stairs hurriedly increased it to 0.20 microampere. Five floor dips increased it to 0.425 microampere, 10 floor dips to 0.625 microampere, and 15 floor dips increased the current to 0.70 microampere. Two minutes after taking the floor dips the current decreased to 0.275 microampere; after four minutes to 0.175 microampere; after six minutes to 0.10 microampere; after eight minutes to 0.10 microampere, and after ten minutes of rest the current had decreased to its original low level of 0.075 microampere. Various kinds of exercises and sports such as football, tennis, baseball, bowling, basketball, handball, boxing, volleyball, skating, badminton, tap

dancing, soccer and wrestling increased the current to a greater or less degree, depending upon the strenuousness of the activity. Exhaustive exercise decreased the current.

Exposure to cold, not sufficient to produce shivering, increased the current to 0.25 microampere. Cold shower bath for two minutes increased it to 0.30 microampere. Hot shower for two minutes increased it to 0.35 microampere.

When a goldfish was placed in water between two platinum disc electrodes with a galvanometer in the circuit a current of low amperage passed from the electrode near the head of the fish to the one near its tail, thus showing the head of the fish to be positive and the tail negative. When the fish became active in swimming the strength of the current increased, as during exercise, in the human. When the fish became quiet the current decreased, as during rest, in the human.

Androgen production in the pregnant and lactating rat. M. W. BURRILL and R. R. GREENE (introduced by F. T. Jung). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Biopsies of ventral prostates from pregnant and lactating rats were studied microscopically. A special strain of rats having a high incidence of female prostates was used. The series of prostates covers the 7th to 22nd day (term) of pregnancy and one to 14 days post partum and represents at least one biopsy for each day during these periods. During the second third of pregnancy the prostates are in a non-functional state, but during the last third, up to and including the 22nd day, the prostates show considerable stimulation consisting of proliferation of the epithelium and occurrence of the light areas which are significant of the functional state. For several days after delivery the prostates appear to regress somewhat, but another period of stimulation appears and persists at least through the 13th day of lactation. The type of stimulation which appears in these prostates is characteristic of androgenic stimulation. The conditions of the prostates demonstrate that, during certain periods of pregnancy and lactation in the rat, androgenic substances are produced in physiologically detectable quantities.

The effect of asphyxia upon plasma volume and protein concentration. D. BAILEY CALVIN. *University of Texas School of Medicine, Galveston.*

Mullin, Dennis and Calvin (Am. J. Physiol. 124: 192, 1938) demonstrated marked increases in blood and plasma potassium in severe asphyxia. These were associated with increased hematocrits, but only slight rises in plasma proteins. Previous work (Calvin *et al.*) has shown quite variable amounts of total circulating plasma protein in response to plasma volume variations.

This communication covers work on the changes in plasma volume, plasma protein (total) and albumin: globulin ratios, brought about by varying degrees of asphyxia. The following results were observed, the degree and kind of response elicited depending upon the depth of asphyxia. From moderate asphyxia to the point of cessation of respiratory movements, plasma volume progressively decreases. Plasma proteins increase in per cent concentration, but there is some decrease in the total amount in circulation, especially in moderately severe asphyxia. Cell volume and per cent plasma protein increases are roughly parallel. There is some fall

in albumin:globulin ratio, due possibly to somewhat greater loss of albumin than of globulin from the circulation.

In severe asphyxia (premortal) there is evidence of massive capillary shock. The plasma volume may be decreased by as much as twenty per cent and although the per cent plasma protein may be slightly above normal, the total results would indicate a marked loss of protein and dye along with water from the vascular system. Further evidence for this is given by cell volume values, which may be elevated as much as thirty per cent, with protein concentration increases below ten per cent. The data agree with Landis' observations on anoxia in isolated capillary studies.

Blood volumes were done using the dye T 1824. There is evidence that some of the dye is lost from the vascular system along with protein in severe asphyxia, since plasma volumes always appeared to be greater after resuscitation than before asphyxia.

In theory it seems possible that loss of consciousness at low oxygen tensions (especially if the condition is attained rapidly) may be due, in part, to capillary anoxia and the resultant sudden decrease in blood volume, yielding serious brain anemia, rather than being due entirely to primary oxygen lack in the brain *per se*.

Studies on the excretion of bromsulfalein in the bile. A. CANTAROW and C. W. WIRTS (by invitation). *Departments of Medicine and Physiology, Jefferson Medical College, and the Laboratory of Biochemistry, Jefferson Hospital, Philadelphia, Pa.*

After intravenous injection of bromsulfalein, the curve of its excretion in liver bile was determined by the Evelyn photoelectric colorimeter. Studies were performed upon bile-fistula dogs (with and without varying degrees of biliary stasis and hepatocellular damage) and human subjects (normal, post-cholecystectomy, various biliary tract disorders, hepatic disease, bile-fistula, etc.).

Under normal conditions, after injecting 2 mgm. of dye per kilogram, the dye appears in the bile within 15 minutes and reaches a maximum concentration (50–532 mgm. per cent) in 45–60 minutes, subsequently falling to a relatively low level at 2 hours and often not disappearing completely for 5–6 hours. Normally, 35–83 per cent of the quantity injected was excreted in the first hour and 61–100 per cent in the first two hours. Since 85–95 per cent of the dye leaves the blood within 5 minutes and all is removed within 30 minutes, two phases of the excretion of bromsulfalein from the body must be recognized: 1, its rapid removal from the blood by, and possibly its storage in the Kupffer and possibly other reticuloendothelial cells; 2, gradual passage of the dye into the hepatic polygonal cells and its excretion in the bile. A third possibility is that temporary storage of the dye occurs in the hepatic rather than in reticuloendothelial cells.

Abnormal excretion of the dye is evidenced by one or more of the following phenomena: 1, delayed removal from the blood; 2, delayed entrance into the bile; 3, delayed attainment of maximum concentration in the bile; 4, prolonged high curve of excretion in the bile; 5, subnormal concentration in bile; 6, abnormally low excretion within 1 or 2 hour periods after injection. The rate of removal of the dye from the blood, utilized in the study of liver function, represents but one phase of the process of its excretion

by that organ and may be normal in the presence of distinct abnormalities in the second or biliary phase of excretion.

The effect of normal urine extract on gastric motility of dogs. E. R. CAPPS (by invitation) and T. L. PATTERSON. *Department of Physiology, Wayne University College of Medicine, Detroit, Mich. and Department of Physiology and Biochemistry, New York Medical College, New York City.*

This study was undertaken to determine the effects of urine extracts on gastric hunger motility. Intravenous administrations of normal, human, female urine extracts in dosages of 2 to 10 mgm. were made on gastric fistula dogs prepared after the method of Carlson, fasted 18 to 24 hours, and contractions registered graphically by the balloon-bromoform manometric method. The gastric tonal characteristics, types of motility and the time relationship of the active and quiescent periods of the empty stomach were observed. Normal gastric motility, as well as that initiated by subcutaneous injections of insulin were employed.

The duration of inhibition by intravenous administrations of urine extracts depended on the dosage and ranged from 10 minutes to 2.5 hours. The latent periods of effective doses were all approximately the same, ranging from 2.5 to 8 minutes, regardless of whether the motility was spontaneous or that induced by insulin.

In three preliminary experiments in which the urine extract was injected subcutaneously, two produced inhibition of gastric motility and the other one was negative. Judging from the two positive reactions, as one might assume, the onset and duration of the inhibition of gastric motility by the subcutaneous route was slower than that produced through the intravenous channels.

These findings would seem to demonstrate conclusively the effectiveness of normal, human, female urine extract as an inhibitant of gastric motility.

On the peroxidatic function of catalase. LOREN CARLSON and GORDON MARSH (introduced by J. H. Bodine). *Department of Zoology, State University of Iowa, Iowa City.*

It has been shown that dilute hydrogen peroxide in oxygenated Ringer's solution increases both the E.M.F. and the respiration of frog's skin. The injection manometer was used for the latter experiments. The consumption of 2×10^{-9} gm. mol. H_2O_2/gm./hr. may cause a simultaneous increase in the oxygen uptake of 80 mm^3/gm./hr. over the control respiration, or roughly an oxygen uptake of 2300 times the peroxide consumed. The ratio of the stimulated oxygen uptake to the peroxide consumed falls to one as the latter increases to 1×10^{-6} gm. mol./gm./hr.

A possible type of mechanism for the stimulating effect of peroxide on the respiration lies in the demonstration by Keilin and Hartree that the peroxide formed by the action of xanthine oxidase on hypoxanthine could be utilized by catalase to oxidize ethyl alcohol, the aldehyde thus formed acting as further substrate for the xanthine oxidase. A similar cyclic oxidation of alcohol by catalase and xanthine oxidase occurred when ethyl or cerium peroxide was used as a source of H_2O_2. The effect was not found with hydrogen peroxide, presumably because of too rapid decomposition.

In the present work the coupled oxidation of alcohol by catalase in the

presence of xanthine oxidase and xanthine is confirmed, using purified preparations of catalase (Sumner and Dounce) and of xanthine oxidase (Ball). The coupled system takes up between two and three times the amount of oxygen consumed by xanthine oxidase and xanthine alone. By use of the injection manometer the addition of 5×10^{-6} gm. mol. H_2O_2 over the course of one hour to 4×10^{-5} gm. mols. ethyl alcohol in the presence of an excess of catalase caused the disappearance of 22 mm^3 oxygen equivalent of peroxide representing the oxidation of 1.96×10^{-6} gm. mols. alcohol. When xanthine oxidase was added to this system the oxygen equivalent taken up was increased to 43 mm^3, or approximately the expectation for the further oxidation of the formed aldehyde to acetic acid.

Mutual cross tolerance between pentobarbital sodium (nembutal) and delvinal sodium [5-ethyl 5-(1-methyl 1-butenyl) barbituric acid] in guinea pigs. EMMETT B. CARMICHAEL. *Department of Physiological Chemistry, School of Medicine, University of Alabama, University.*

The existence of cross tolerance has been established between the two barbituric acid derivatives: 1, pentobarbital sodium, and 2, delvinal sodium.

Young guinea pigs were used and all injections were made intraperitoneally. A single large dose of one of the above drugs was used and after a few days it was followed by a series of semi-weekly doses of the other drug for a period of three weeks.

Two series of experiments were made, using pentobarbital sodium as the initial drug: 1, sixteen guinea pigs received 20 mgm./kgm., and 2, thirteen guinea pigs received 50 mgm./kgm. The animals in each of these series received semi-weekly doses of 40 mgm./kgm. of delvinal sodium for three weeks. The average length of hypnosis following the delvinal sodium was reduced from that produced by the same size dose of this drug on normal guinea pigs of about the same weight. The average length of hypnosis for normal guinea pigs was 192 minutes and following the 50 mgm. dose of pentobarbital, the average length of hypnosis was reduced to 169 minutes.

A group of 16 guinea pigs that survived a single dose of 70 to 85 mgm. of delvinal sodium/kgm. were given semi-weekly doses of 20 mgm. of pentobarbital sodium/kgm. for three weeks. A definite tolerance was also produced in this series. This was shown by the fact that the length of hypnosis following the first dose of the pentobarbital sodium was markedly less than it was for normal controls with the same size dose/kgm. The average length of hypnosis in the control experiments with a dose of 20 mgm. of pentobarbital sodium/kgm. was 93 minutes while the duration of the hypnosis, following a large dose of delvinal sodium, was 49 minutes

The relation of rhythm to the force of contraction of mammalian cardiac muscle. MCKEEN CATTELL and HARRY GOLD (by invitation). *Department of Pharmacology, Cornell University Medical College, New York City.*

The experiments of Woodworth (Am. J. Physiol. **8:** 213, 1902) which demonstrate that, in the isolated apex of the dog heart, the extent of shortening is directly related to the frequency or inversely to the spacing between responses, has not received the attention its importance deserves. We have confirmed and extended these observations on isolated papillary muscles from the right side of the cat heart, using the isometric response. This

preparation exhibits a striking "treppa" when stimulated at a regular frequency after a period of rest. It shows a several-fold increase in the force of contraction when the rate is changed from a slow to a fast one, and a decrease in force when the rate is changed in the reverse direction. These changes in force develop gradually. The first few contractions in the transition from a low to a high frequency are smaller than the previous ones, and conversely, the first few contractions in the transition from a high to a low frequency are larger than the previous rapid beats and subsequent slow beats.

A single extra stimulus applied sufficiently close to one in the regular series causes a large increase in the next regular response. The closer the paired stimuli the greater the succeeding response; thus a maximum augmentation occurs when the extra stimulus falls well within the relative refractory period, so that of itself it produces only a minimal contraction. The influence of the closely spaced extra beat is only gradually dissipated and after five minutes with the muscle at rest is still largely present.

An analysis of the records leads to the conclusion that the factor which is responsible for the increased force of contraction during the treppa and at higher frequencies of stimulation or following a single extra stimulus is the rhythm or spacing between stimuli rather than the increased activity *per se*.

Micromanipulative studies on vascular responses to localized micro-injury. ROBERT CHAMBERS. *Washington Square College, New York University, New York City.*

The experiments were made on the mesentery and tongue of the frog immobilized by careful destruction of the forebrain and after the ensuing transitory hyperemia had passed off. Micro-injuries of graded intensity were performed by inserting a microneedle and moving it about in the tissue. Care was taken to produce the injury remote from neighboring arterioles and venules and not to tear the capillaries. During normal circulation the arterioles exhibit a slow periodic dilatation and contraction. The constrictor effect is undoubtedly of sympathetic nervous origin. The local origin of the dilator effect was corroborated by experiments with micro-injury. The venules, on the other hand, maintain a constant caliber, the variation in the amount of blood from the arterioles being adjusted mainly by the number of the capillaries involved. Experimentally produced variations in blood pressure were compensated for by a corresponding contraction and dilatation of the arterioles, the venules maintaining a constant diameter.

On the other hand, the initial effect of a micro-injury was found to be a dilatation of the venule. This was followed by a preliminary slowing of the stream in the capillaries, then a loss of the rhythmic contractions of the arterioles which, nevertheless, still responded to prodding. Subsequently there was a loss in tone of all the vascular vessels. All the capillaries then become filled and, from the excessive loss of fluid, become choked with red cells. Following the trauma induced, recovery set in about 70 to 90 minutes after the injury. The initial step observed was a loosening of the clumped cells in the venous capillaries. With the resumption of blood flow there was a narrowing of the venules and a return of the rhythmic contraction of the arterioles. The initial involvement of the venules after injury

may be explained by a spread, downstream, of a highly diffusible substance emanating from the site of injury. Subsequently, this substance spreads throughout and also affects directly the musculature of the arterioles. Alterations in the walls of the vessels were noted by the diffusibility of dyes. A check on the spread of a tone-lowering substance was obtained by rerouting venous blood back through the capillary network.

Equilibrium and kinetic studies of cyanide inhibition of peroxidase. BRITTON CHANCE (introduced by A. N. Richards). *Johnson Foundation, University of Pennsylvania, Philadelphia.*

Spectroscopic measurement of the rapid reactions of enzyme and substrate and enzyme and inhibitor correlated with the corresponding equilibrium studies have permitted a detailed study of the mechanism of cyanide inhibition of the hematin enzyme peroxidase. In the absence of substrate the equilibrium constant for peroxidase and cyanide is 4×10^{-6} and one mole of inhibitor combines with one mole of the enzyme. Although the rate of this reaction is rather rapid ($k_2 = 8 \times 10^4$ liters $\times$ mol$^{-1} \times$ sec^{-1}), it is only one hundredth the rate of combination of enzyme and substrate. There is then competition between the inhibitor and substrate for the enzyme even at very low substrate concentration (4×10^{-6} M/L). This is demonstrated by studies on the effect of cyanide on the overall enzyme action as determined by the rate of production of malachite green. For a particular substrate concentration the calculated equilibrium constant for peroxidase and cyanide is increased fivefold. These data, however, confirm a one to one proportionality between hematin iron and cyanide. Studies of the kinetics of the enzyme-substrate compound in the presence of inhibitor show under certain conditions the practically uninhibited operation of the enzyme-substrate compound followed by the combination of enzyme and inhibitor only after the substrate has been nearly exhausted.

Experiments on the combination of catalase and cyanide indicate that the rate constant ($k_2 = 5 \times 10^5$ liters $\times$ mol$^{-1} \times$ sec^{-1}) is likewise considerably smaller than the calculated minimum rate (Haldane) for the combination of enzyme and substrate.

The competition between inhibitor and substrate for the enzyme and the resultant effect upon the equilibrium constant for the enzyme-inhibitor compound as determined by measurements of overall enzyme activity are clearly demonstrated.

The absorption spectrum of luciferin solutions during luminescent and non-luminescent oxidation. AURIN M. CHASE. *Physiological Laboratory, Princeton University, Princeton, N. J.* (Read by title.)

Absorption spectra of concentrated aqueous solutions of purified Cypridina luciferin of pH 6.8 have been measured with the Hardy recording spectrophotometer. Solutions of freshly dissolved luciferin show an absorption maximum at about 430 mμ. During oxidation by air, in absence of luciferase, this maximum shifts to 470 mμ, and this second maximum then disappears, leaving an almost colorless solution.

The absorption spectrum has also been measured during luminescent oxidation of luciferin in the presence of luciferase. Here exactly the same sequence of changes occurs as in spontaneous non-luminescent oxidation

by air. However, the rate of these changes is at least one hundred times as great when luciferase is present. No difference other than one of rate can be detected between the luminescent oxidation with luciferase and the non-luminescent oxidation by air, so far as the color changes of the solution are concerned.

The absorption spectrum of a freshly-prepared aqueous luciferin solution is the same whether the pH be 6.8 or 5.0. However, during spontaneous, non-luminescent oxidation the changes in the absorption spectrum are much less rapid at the acid pH and, in addition to this difference, absorption remains greater at the short wavelengths. This indicates that in the oxidation of luciferin by air a compound is produced whose color depends upon the pH of the solution.

If luciferin solutions of different degrees of purity (so far as color is concerned) are measured, it is found that the concentration of luciferin present, as determined by measurements of total luminescence, is proportional, not to the total color of the solution, but to the labile color. This is strong evidence that these changes in the absorption spectrum represent luciferin itself.

Gastric secretion after treatment with the histamine antagonist, thymoxyethyldiethylamine. ORVILLE CHICKERING (by invitation) and EARL R. LOEW. *Wayne University College of Medicine, Detroit, Mich.*

Thymoxyethyldiethylamine (929F) has been reported to prevent histamine from causing bronchioconstriction and contraction of guinea pig ileal strips. Furthermore, it has been claimed that treatment with 929F prevents fatal anaphylactic shock in guinea pigs and protects such animals against several minimal lethal doses of histamine.

Hence, experiments were performed to determine whether 929F inhibits the secretion of gastric juice induced by subcutaneous injections (0.5 mgm.) of histamine. The double histamine test was carried out upon dogs provided with Heidenhain pouches. Control data consisted in measurements of volume of gastric juice and total, combined, and free acid secreted during a period of 2 hours following the histamine stimulus. The dogs were then given 10 mgm. of 929F per kgm. subcutaneously, and one-half hour later the same histamine stimulus was given. Gastric secretion after treatment with 929F was no less than that which occurred during the control periods. The doses of 929F administered produced no untoward reactions. Larger doses were not employed since retching, vomiting, and defecation usually resulted.

The results indicate that 929F, in non-toxic doses, does not depress or alter the composition of the gastric juice induced by subcutaneous injections of histamine.

The effect of lipocaic on essential xanthomatosis. DWIGHT E. CLARK (by invitation), ORMAND C. JULIAN (by invitation), CORNELIUS W. VERMEULEN (by invitation), J. GARROTT ALLEN (by invitation) and LESTER R. DRAGSTEDT. *Department of Surgery, The University of Chicago, Chicago, Ill.*

Four cases of xanthoma, all of which showed a very marked elevation in the blood lipids, were treated with lipocaic. In 3 of the cases the lesions manifested themselves in the skin and in the other case the xanthomas were

connected with tendons. In all 4 cases the blood lipids were reduced to normal levels following the administration of lipocaic. Two of the cases in which the lesions were in the skin cleared completely, while the other was markedly improved. In the case where the xanthomas were associated with the tendons, the pathological lesions were excised. Lipocaic was administered to this case as a prophylaxis against recurrence.

This study substantiates the clinical inference that xanthomas are associated with a disturbance of lipid metabolism and lipocaic may prove to be a valuable adjunct in the treatment of this disease. Also it adds to the evidence that lipocaic plays an essential role in lipid metabolism.

Changes in optical rotation of fibrinogen with gel formation. JANET H. CLARK. *University of Rochester, Rochester, N. Y.*

Proteins are known to exhibit changes in specific rotation as the result of changes in hydrogen ion concentration, denaturation, or with sol $\rightleftarrows$ gel transformations. A study was made of the specific rotation of fibrinogen setting to a gel on addition of thrombin. Fibrinogen and thrombin are both levo-rotatory but when thrombin is added to a solution of fibrinogen the immediate result is the formation of a dextro-rotatory compound. When a clot begins to form this changes to a levo-rotation, probably due to structural asymmetry in the gel, which increases rapidly reaching values as high as -280 deg./dm.

Prolonged action of desoxycorticosterone acetate.[1] C. F. CODE, R. A. GREGORY (by invitation), R. E. LEWIS (by invitation) and F. J. KOTTKE (by invitation). *Department of Physiology, University of Minnesota and Mayo Foundation, Rochester.*

It has been recently demonstrated that the action of a single injection of histamine may be greatly prolonged by suspending the histamine particles in a beeswax-mineral oil mixture (Code and Varco, Proc. Soc. Exper. Biol. and Med. **44:** 475, 1940). We have found that the action of desoxycorticosterone acetate may also be prolonged by suspension in the beeswax mixture. In order to determine the period of action of a single injection of desoxycorticosterone acetate the daily output of chloride in the urine was followed in adrenalectomized dogs on diets of fixed amounts of meat and milk. The level of urea in the blood was also measured for an additional check on the action of the desoxycorticosterone acetate. The beeswax mixture was prepared by mixing 1 part hot beeswax with finely ground desoxycorticosterone acetate and diluting with between 4 and 5 parts hot mineral oil. While still molten the mixture was drawn into a 1 cc. tuberculin syringe. When it cooled to room temperature it could be ejected through a 22 gauge needle. The period of action of this mixture was compared to the period of action of similar doses of desoxycorticosterone acetate dissolved in sesame oil. In all instances the desoxycorticosterone acetate was injected subcutaneously.

It was found that 25 mgm. of desoxycorticosterone dissolved in sesame oil maintained the adrenalectomized dogs for 2 or 3 days while the same

[1] Part of the expense of this research was defrayed by a grant from the Committee on Scientific Research of the American Medical Association. The major part of the desoxycorticosterone acetate used in this investigation was provided through the kindness of Ciba Pharmaceutical Products.

dose placed in the beeswax mixture was effective for 8 to 13 days. With doses of 100 mgm. the contrast was even more marked. Dissolved in sesame oil, 100 mgm. desoxycorticosterone was generally effective for only 5 days while in the beeswax mixture the period of action of this dose was extended to one month or longer.

Fractionation of the steroids from human postpartum urine. SAUL L. COHEN (introduced by Frank A. Hartman). *Department of Physiology, The Ohio State University, Columbus.*

A 50 l. batch of urine was collected from humans during the first 24 hours after parturition. The ether soluble portion of this urine was obtained both before and after treatment with strong acid, in order to obtain the "free" and "combined" compounds respectively. The ether solutions were extracted first, with sodium carbonate and then with sodium hydroxide solutions to yield neutral, acidic and phenolic fractions. The extract containing the free neutral compounds has been further fractionated as follows: The ketonic and non-ketonic substances were separated by means of Girard's reagent. The non-ketonic fraction was adsorbed on a column of active aluminum oxide, and the column was then subjected to a series of elutions with solvents of increasing polarity. These elutions have yielded a light colorless oil and five distinct crystalline compounds, two of which have been identified as cholesterol and pregnandiol respectively. Some properties of the other three compounds with melting points of 46°, 72° and 108° (uncorrected) respectively will be discussed.

The beta-glycerophosphatase activity of the mammalian central nervous system. DAVID J. COHN and IRVING KAPLAN (introduced by Heinrich Necheles). *Department of Biochemistry, Michael Reese Hospital, Chicago, Ill.*

The beta-glycerophosphate hydrolyzing activity of different parts of the central nervous system of dogs, rhesus monkeys, rats and rabbits was found to differ with the location of the tissue. Minced fresh tissue was used as the enzymic material, and the activity was determined by a modification of the method of A. Bodansky. The results showed that the glycerophosphatase activity was greatest in grey matter, least in white matter, and intermediate in mixed tissues. The average activities for seven adult dogs (expressed in milligrams of inorganic phosphorus liberated per gram of fresh tissue in one hour at 37.5°C.) were: cerebral cortex, 1.01; striatal grey matter, 0.77; mixed cerebral tissue, 0.68; thalamus, 0.66; cerebellum, 0.61; pons and medulla (combined), 0.47; spinal cord, 0.26; white matter, 0.28. The average values for 5 adult rabbits were: cerebral cortex, 0.31; basal nuclei, 0.25; cerebellum, 0.24; mixed cerebral tissue, 0.20; pons and medulla, 0.20; and spinal cord, 0.07. Values for the rhesus monkey were approximately midway between those of the dog and rabbit. In other words, there are definite species differences.

Activity was found to decrease with the age of the animal. The activity of different parts of the central nervous system of young puppies was greater than that of the same tissues of the adult dog, particularly in the stem and spinal cord. The activity of the whole brain of new born rabbits was between 2 and 3 times as high as that of the cerebral cortex of adult rabbits. Similarly, brains of young rats were more active than

those of adult rats. These results indicate that phosphatase may play an important part in the synthesis and deposition of phospholipid in the infant brain, where the rate of such deposition has been shown to be greater than in the adult.

Observations on the permeability of mammalian cells to cations.[1] WALDO E. COHN (introduced by Joseph C. Aub). *Division of Biochemistry, University of California, Berkeley, and the Collis P. Huntington Memorial Hospital, Harvard University, Boston, Mass.* (Read by title.)

The artificial radioactive isotopes of sodium and potassium afford a direct method for measuring the permeability of cells to these ions. After intravenous injection of Na^{24} or K^{42}, in the form of NaCl or KCl solution, the radioactive isotope disappears rapidly from the plasma, reaching a relatively constant level within two hours. From the value of this plateau level, the amount of radioactivity which has left the extracellular phase can be calculated, while radioactive assay of various tissues offers a direct measure of relative permeabilities.

From studies of this kind on normal dogs, it is concluded that Na^{24} distributes itself throughout extracellular fluid in about 100 minutes, no appreciable amount entering cells with the exception of the erythrocytes. These undergo a slow exchange of sodium, requiring about 12 hours for half-completion. Injected K^{42}, however, not only is distributed throughout the extracellular fluid within this time, but also, in a 10-fold greater concentration, throughout the intracellular fluid. It is calculated that this distribution of K^{42} represents an exchange between intracellular and extracellular K involving 40 per cent of the former.

The intraperitoneal administration of these isotopes to rats and guinea pigs leads to similar results, except that the exchange of erythrocyte potassium is not nearly as complete as in the dog.

In humans, the initial rapid exchange of K^{42} for cellular K is less, involving about 20 per cent of the intracellular potassium. The K^{42} concentration of the erythrocytes rapidly approaches that of the plasma, which represents only a very small exchange of their total K.

These results are interpreted as representing an exchange of potassium between intra- and extra-cellular fluids. There is evidence to support the view that cellular K exists in two forms, one readily exchangeable, the other slowly so. The entry of K^{42} seems to be related to the K content of the individual tissue. In all species examined, the amount of K^{42} that had left the extracellular phase was many times that remaining in it.

Longitudinal impedance of the squid giant axon. KENNETH S. COLE and RICHARD F. BAKER (by invitation). *Department of Physiology, College of Physicians and Surgeons, Columbia University, New York City.*

Longitudinal alternating current impedance measurements have been made on the squid giant axon over the frequency range from 30 cycles per second to 200 kilocycles per second. Large sea water electrodes were used and the inter-electrode length was immersed in oil. The impedance at high frequencies was approximately as predicted theoretically on the basis of the poorly conducting dielectric characteristics of the membrane

[1] The radioactive isotopes used in this investigation were supplied by the cyclotrons of the University of California and Harvard University.

previously determined. For the large majority of the axons, the impedance reached a maximum at a low frequency and the reactance then vanished at a frequency between 150 and 300 cycles per second. Below this frequency the reactance was inductive, reaching a maximum and then approaching zero as the frequency was further decreased.

The inductive reactance is a property of the axon and requires that it contain an inductive structure. The variation of the impedance with the interpolar distance indicates that the inductance is in the membrane. The impedance characteristics of the membrane as calculated from the measured longitudinal impedance of the axon may be expressed by an equivalent membrane circuit containing inductance, capacity and resistance. For a square centimeter of membrane the capacity of one microfarad with dielectric loss is shunted by the series combination of a resistance of four hundred ohms and an inductance of one-fifth henry.

The electrical impedance of single frog eggs. KENNETH S. COLE and RITA GUTTMAN (by invitation). *Department of Physiology, College of Physicians and Surgeons, Columbia University, New York City.* (Read by title.)

The electrical impedance of single frog eggs was investigated with an alternating current Wheatstone bridge over the frequency range from 0.05 kc. to 10 kc. Measurements were made on 19 eggs in either spring water or a 10 per cent amphibian Ringer's solution—both of which permit normal development.

The resistance of the vitelline membrane was found to be negligible. The plasma membrane resistance averaged 170 ohm cm^2, varying from 65 to 570 ohm cm^2. This resistance lies in the range of membrane resistances found for the squid axon, frog sartorius muscle, Valonia and Halicystis, and suggests that the ion permeabilities of these membranes are at least comparable. The membrane capacity averaged 2.0 microfarads/cm^2, varying from 1.3 to 2.9 microfarads/cm^2, which is in good agreement with the values generally obtained for membrane capacities in a large variety of cells. The specific resistance of the egg interior averaged 570 ohm cm, varying from 320 to 720 ohm cm. The phase angle of the membrane averaged 86°, varying from 81° to 89.5°.

There is no certain change in membrane resistance or membrane capacity on fertilization, and also the specific resistance of the cell interior and phase angle of the membrane do not change upon fertilization. The spontaneous rhythmical impedance changes described by Hubbard and Rothschild (Proc. Roy. Soc. London, Series B **127:** 510, 1939) for the trout egg were not found in the frog egg.

Effects of direct chemical and electrical stimulation of the respiratory center. JULIUS H. COMROE, JR. (introduced by C. F. Schmidt). *Laboratory of Pharmacology, University of Pennsylvania, Philadelphia.*

In 65 cats, anesthetized with nembutal or decerebrated under evipal, the medulla and pons were explored with a hollow steel needle (0.5 mm. in diameter, inserted in the Horsley-Clarke instrument) which served both as injecting needle and electrode, a copper rod in the rectum being the indifferent electrode.

Acid solutions (N/1000 to N/10 HCl or lactic acid) were injected 337

times in 1.0 to 5.0 cu. mm. amounts, from 2 mm. below to 5 mm. above the obex, with these results: in 8 instances, barely perceptible stimulation of respiration occurred; in 3, slight hyperpnea resulted; in one, an inspiratory apnea developed. Following the remaining 325 injections (96 per cent) no change or respiratory depression was observed. Acid solutions of CO_2 in saline (70, 300, or 700 mm. tension) likewise failed to stimulate respiration.

A striking contrast was observed when solutions were used containing 1.31 to 1.57 per cent $NaHCO_3$ in distilled water, buffered with CO_2 to pH 7.4, CO_2 tension being 250–300 mm. (though similar results were obtained with CO_2 tension as low as 100 mm.). Of 740 injections into pons and medulla (6 mm. below to 16 mm. above obex), 214 (29 per cent) resulted in immediate mild to vigorous hyperpnea. Of 387 injections in medulla alone (1 mm. below to 4 mm. above obex) 40 per cent were positive; in the center of this region (corresponding to the reticular formation) 80 per cent positive responses were elicited. Although occasionally tonically-maintained inspiration or expiration was noted, similar to that observed by Pitts *et al* following electrical stimulation of this region, the characteristic response was increase in respiratory rate and depth. Control injections of distilled water, hyper- and hypotonic saline were negative though bicarbonate solutions, pH 7.7, elicited positive responses. Vasomotor responses in this region were inconsistent. The results of electrical stimulation were in general agreement with those reported by Pitts.

Conclusions. 1. Responses to chemical stimulation of the medulla localize the respiratory center to the medullary reticular formation. The importance of the hydrogen ion per se as the characteristic stimulus to the respiratory center is open to serious question.

The effect of postural changes on blood pressure in the rabbit. Ruth E. Conklin and Virginia C. Dewey (by invitation). *Vassar College, Poughkeepsie, N. Y.* (Read by title.)

A study was made of the part played by presso-receptors of the carotid sinus and aortic region in the reflex control of blood pressure in the rabbit. This animal was chosen because of its lax abdominal wall, making it peculiarly ill-adapted to posture changes, and because it possesses a depressor nerve separate from the vagus.

Normal, adult rabbits, anesthetized with urethane, were used. The animals were fixed on a tipping board which could be tipped to the foot-down position before and after cutting the vagi or depressor nerves or denervating the carotid sinuses. Blood pressure was recorded by a modified Hürthle manometer. Twenty-eight successful tipping experiments were done, with tipping from 15° to 45°.

The typical response was a rapid fall in blood pressure, followed, while the animal was still tipped, by a slower rise to or toward the pre-tipping level. The only difference between the response of the control animals and those deprived of vagi, aortic arch receptors, carotid sinus receptors or all three was a slightly smaller fall in blood pressure after destruction of presso-receptor pathways than before. The control animals averaged 61 per cent of their initial blood pressure when they were tipped 30°. Animals deprived of vagi, depressors and carotid sinuses averaged 78 per cent of their initial blood pressure at 30°. Since the power of compensa-

tion is not lost in animals deprived of these sources of receptor impulses, other areas must be responsible for compensatory reflexes. Further work is now in progress along this line.

Variable frequency stimulator and recorder. HAROLD V. CONNERTY and WALTER H. JOHNSON (introduced by Reginald A. Cutting). *Georgetown University School of Medicine, Washington, D. C.* (Demonstration.)

An electrical apparatus composed of standard radio parts has been devised for the stimulation of nerves and muscles at accurately controlled frequencies and intensities. Stimuli can be produced at rates between one shock every six seconds and close to twenty-five hundred per second. The voltage of the stimulating current at any one frequency can be independently controlled. An outlet is provided for a signal magnet which works in complete unision with the stimulating current; the blinking of an "electric eye" also provides an indication of the frequency.

The electrical characteristics of the instrument will be demonstrated by means of an oscilloscope. This will show that each stimulation is by direct current of extremely short duration, that the voltages of the stimuli at any one frequency are absolutely uniform, and that output voltages can be varied at will without effecting the frequency.

Kymograms of the responses of a frog's gastrocnemius muscle to different intensities and frequencies will be on exhibit.

The action of inorganic salts and cardiac glucosides on the turtle heart. HELEN C. COOMBS and F. H. PIKE. *Department of Industrial Hygiene of the Delamar Institute of Public Health, Columbia University.* (Read by title.)

Perfusion of the heart with varying concentrations of inorganic salts has been done on more than sixty turtles. The following observations are submitted at this time.

In late spring and summer, when the heart is perfused with Ringer's solution containing an excess of potassium it stops in diastole, as noted by previous investigators. One cat unit of cardiac glucoside (strophanthin or digitalis) added to control Ringer gave only slight effects. When the concentration of potassium was increased to ten times that of the control Ringer, addition of the same amount of glucoside as before was usually followed by the cessation of the beat in the position of systole. Sometimes a still greater concentration of potassium (M/10) was necessary for this effect.

The effects were more variable in fall and winter and suggest a seasonal change in the heart. When the concentration of potassium is increased the heart often stops in systole. Less potassium is usually sufficient to stop the heart in systole when a cardiac glucoside is added than in summer. Great sensitivity of the heart to potassium is apparently related to the degree of infestation of the intestines by nematode or acanthocephalid parasites and a yellowish-brown liver. This relationship is being checked further. It is possible that failure of the turtle's heart in student laboratories may be related to general systemic changes.

Certain hearts sensitive to excess potassium are also sensitive to lack of calcium. In the absence of calcium, the heart soon shows irregularities of rhythm with a tendency to shorten and stop in systole. Also, in the case

of the heart which is sensitive to excess of potassium, even to the point of stopping in systole, addition of calcium to the perfusion fluid is followed by relaxation of the heart.

The action of excess potassium is, in general, more marked in winter than in summer, and after the heart has been perfused with Ringer minus calcium.

Tension in antagonistic muscles in voluntary and reflex movement. JOHN P. COOPER and HERMAN KABAT (introduced by M. B. Visscher). *Department of Physiology, University of Minnesota, Minneapolis.*

The cut tendons of the anterior tibial and soleus muscles were connected to tambours for simultaneous recording of tension in the antagonistic muscles. The animal was under light ether anaesthesia.

In flexion, three types of responses were observed in the extensor muscle. Frequently, the tonus of the extensor increased, with the maximal tension in the extensor small compared to that in the flexor (co-contraction). In other instances, the tonus of the extensor decreased (reciprocal innervation). In still other cases, there was no change in tonus of the extensor as the flexor contracted.

In spontaneous flexion, 55 per cent of the responses showed co-contraction and 32 per cent showed reciprocal innervation. This is in agreement with the work of Tilney and Pike (1925), who found almost exclusively co-contraction during stimulation of the motor cortex.

Co-contraction also predominated in reflex flexion. On stimulation of the foot pad, 78 per cent of the responses showed co-contraction and only 10 per cent reciprocal innervation.

In another series of experiments, the spinal cord was transected at the first lumbar segment and the flexion reflex studied 3 hours later. When the foot pad was stimulated, reciprocal innervation was found to be more frequent (40 per cent) and co-contraction less frequent (26 per cent).

In another group of cats, the tonus of the extensor was increased by increased stretch of the muscle or by stimulation of the crossed extension reflex. When extensor tonus was low, the flexion reflex showed 86 per cent co-contraction and 7 per cent reciprocal innervation. When extensor tonus was high, the flexion reflex showed 13 per cent co-contraction and 87 per cent reciprocal innervation.

This preliminary investigation indicates that when the tonus in the antagonist is low or moderate, co-contraction is the most frequent response in reflex or voluntary movement. When the tonus of the antagonist is high or after transection of the spinal cord, reciprocal innervation becomes most frequent. These factors may, perhaps, account for the observation of Sherrington that reciprocal innervation is characteristic of the action of antagonistic muscles. Sherrington states (1910) that a high tonus in the antagonist is necessary for the demonstration of reciprocal innervation.

Bleeding time in men. ALFRED L. COPLEY[1] and JOSEPH J. LALICH[2] (introduced by O. O. Stoland). *Hixon Laboratory for Medical Research, University of Kansas, School of Medicine, Kansas City.*

[1] Aided by a grant from the Dazian Foundation for Medical Research.
[2] George A. Breon Fellow in Experimental Medicine.

There are several techniques in use for the determination of bleeding time in men. We eliminated variations of temperature, and of venous hemodynamic pressure, and attempted to produce a uniform prick wound, in order to obtain more constant conditions.

The principle of Doettl and Ripke of bleeding into fluid (1938) was adopted for capillary blood. A constant temperature bath (38 to 39°C.) capable of heating 200 cc. of isotonic saline with illumination to observe the bleeding, was constructed. The third or fourth finger was cleaned with alcohol, and immersed for two minutes into the bath. The heated phalanx was removed temporarily while a prick wound was inflicted with an automatic lancet (blade dimensions 2 x 6 mm.), after which the finger was immediately immersed into the bath. The bleeding time was measured with a stop watch from the moment the wound was inflicted, until the blood flow stopped.

A series of 222 tests were done on 114 persons composed of the clinic staff, medical students, and ambulatory clinic patients. The range of bleeding times was between 19.6 and 325.0 seconds, the average was 95 seconds; 182 values were between 45 and 165 seconds; 20 values were less than 45 seconds, and 20 values were more than 165 seconds. With strong illumination, a colorless flow which we believe is tissue fluid, was observed after the red flow had stopped. In our determinations we used the cessation of the red flow as the end point of bleeding time. Room temperature prolonged both kinds of flow.

Bleeding time in normal and heparinized mice. ALFRED L. COPLEY[1] and JOSEPH J. LALICH[2] (introduced by O. O. Stoland). *Hixon Laboratory for Medical Research, University of Kansas School of Medicine, Kansas City.* (Read by title.)

The hemostatic function of the skin is measured by the bleeding time. Doettl and Ripke (*Medicine in its chemical aspects* **3**: 252, 1938, Bayer, Leverkusen, Germany) described a test in mice which we used in this study. We differentiated various strength of flows, and an arterial flow from a venous flow by its pulsation. Two hundred and ninety-five determinations in 106 mice ranging between 15.4 to 220.3 seconds had an average bleeding time of 53.8 seconds. Unlike Doettl and Ripke, we regard close checking on following days to be within 30 seconds.

The effect of four preparations of heparin was studied on the bleeding time. Heparin (Connaught Laboratories, 110 units per mgm.) in large doses (1000 units per 20 grams weight injected subcutaneously) had no toxic effect. There is a definite relationship between the units of heparin and the prolongation of clotting time. Such a relation to bleeding time does not exist in smaller doses (5 to 100 units), whereas in excessive doses (200 to 1000 units per 20 grams weight), and then only in some instances, was there an increase of both the bleeding and coagulation time. Repeated prickings in some excessively heparinized mice produced a prolonged bleeding time after several normal values, an observation never seen in normal mice.

We believe that there is a factor present in mice which neutralizes the action of heparin and that in contrast to coagulation time only excessive doses of heparin prolong the bleeding time.

[1] Aided by a grant from the Dazian Foundation for Medical Research.
[2] George A. Breon Fellow in Experimental Medicine.

The effect of heparin on the platelet count in dogs and mice. ALFRED L. COPLEY[1] and TOM P. ROBB (introduced by O. O. Stoland). *Hixon Laboratory for Medical Research, University of Kansas School of Medicine, Kansas City.* (Read by title.)

The Vilarino-Pimentel method (Klin. Wchnschr. **18:** 1253, 1939) was used to make platelet counts on venous blood of dogs and heart blood of mice.

In vitro: Heparin (Connaught Laboratories, 100 units per mgm.) decreased the platelet count directly proportional to concentration but not to action time. In one instance platelets entirely disappeared within 15 minutes after addition of 25 units of heparin. Three more cases having the same dose and action time showed 44, 67, and 71 per cent decrease. It seems that certain platelets resist the action of heparin.

In vivo: Intravenous injection of 100 and 200 units per kilogram weight into dogs was followed by decreased counts in almost all cases. No relationship between dose and decrease of count was seen. Twenty-five mice injected subcutaneously with heparin 5 to 1000 units per 20 grams weight showed most frequently counts below the range established in 22 normal mice. Five mice having excessive hemorrhage and long bleeding time showed normal and increased counts. No relationship between bleeding time, heparin dosage and platelet count was seen.

We suggest that there exist in animals mechanisms which maintain the amount of platelets. We believe heparin has disintegrating effects upon platelets.

A new direct method of counting the blood platelets. ALFRED L. COPLEY[1] and TOM P. ROBB (introduced by O. O. Stoland). *Hixon Laboratory for Medical Research, University of Kansas, School of Medicine, Kansas City.* (Read by title.)

Into a syringe containing 1 cc. of modified Aynaud solution (sodium chloride 0.75 per cent, sodium citrate 3.8 per cent, formaldehyde 3.7 per cent), 1 cc. of venous blood is drawn. The blood and solution are mixed and the syringe is emptied into a clean dry test tube. This first dilution may be kept stoppered at room temperature for 24 hours. At the time of the count the tube is inverted 10 times and 0.5 cc. of the mixture is added to 12 cc. of 3.8 per cent sodium citrate. The second dilution is mixed and 0.5 cc. is added to 0.5 cc. of 0.2 per cent brilliant cresyl blue in 3.8 per cent sodium citrate. The mixed final dilution is placed in a counting chamber. The red blood cells will fade after 10 minutes, and the platelets will appear as dark blue bodies. A few red blood cells that take the stain are differentiated by size and shape. These counts compare with the Vilariño-Pimentel method (1939) ± 2.5 per cent in 11 dogs and 3 humans.

The method is simple, requires inexpensive equipment, and is convenient for transporting the blood mixture and deferring the count. The platelets are preserved from disintegration by formaldehyde upon withdrawal. Accuracy is also increased because of the large volume of blood and the low final dilution used, and because the errors in the use of cutaneous blood are avoided.

Effects of renal extracts containing "angiotonin-inhibitor" on renal blood flow and function in normal and hypertensive dogs and human beings.

[1] Aided by a grant from the Dazian Foundation for Medical Research.

A. C. Corcoran, K. G. Kohlstaedt (by invitation) and Irvine H. Page. *Lilly Laboratory for Clinical Research, Indianapelis City Hospital, Indianapolis, Ind.*

Renal blood flow and function was observed in normal and experimentally hypertensive dogs and in hypertensive human beings before and during administration of renal extracts. The measurements were made from the clearances of diodrast, phenol red, and inulin.

Administration of extracts containing angiotonin-inhibitor usually increased renal blood flow and always caused efferent arteriolar vasodilation. The effect was most intense in normal dogs. In these there occurred no significant change of blood pressure unless the amount given was very large. The renal action of the extracts was associated with decreased arterial pressure in both hypertensive dogs and human beings. On three occasions, mean arterial pressure of hypertensive dogs was reduced to normotensive levels, and renal blood flow, which had at first increased as the pressure fell to about 140 mm. Hg, fell precipitously. Renal blood flow returned to higher levels as arterial pressure later increased.

In a few patients suffering from malignant hypertension, renal blood flow did not increase greatly as arterial pressure fell, although evidences of efferent arteriolar vasoconstriction all but disappeared. Renal blood flow in no case decreased during the use of these extracts in patients suffering from hypertension. It is therefore suggested that renal ischemia which sometimes occurred in experimentally hypertensive dogs at low arterial pressures was the result of mechanical interference with the flow, whereas, in human beings, the obstruction to renal blood flow was largely due to vasoconstriction.

These observations lead to the conclusion that the renal extracts were not only anti-pressor, but also inhibited the renal action of the humoral pressor substance, angiotonin. The data also imply that the renal ischemia of experimental renal hypertension is partly due to intrarenal vasoconstriction as well as to mechanical interference with renal circulation. The observations made in normal dogs suggest that the renal vaso-pressor system may play some part in the normal regulation of renal blood flow.

Renal blood flow in experimental hypertension due to constriction of the renal artery. A. C. Corcoran and Irvine H. Page. *Lilly Laboratory for Clinical Research, Indianapolis City Hospital Indianapolis, Ind.* (Read by title.)

Reduction of renal blood flow will result from constriction of the renal artery if mean arterial pressure has been reduced distal to the point of clamping. If the clamp be tightened only enough to dampen the pulse, and if systemic arterial pressure is somewhat increased, intra-renal arterial pressure may be unchanged and renal blood flow maintained at a normal level. Secondary reduction of renal blood flow may occur from constriction of the efferent arterioles by renin and angiotonin, humoral agents of renal hypertension. However, if the systemic hypertension is sufficient to result in increased intra-renal arterial pressure, the resistance due to intra-renal vasoconstriction will be overcome, and renal ischemia will not occur. This balance of factors will not usually be obtained unless special efforts are made.

Observations previously reported (Corcoran and Page, Am. J. Physiol. **122**: 43P, 1938) have shown that hypertension in uninephrectomized dogs due to compression of the renal artery may occur without depression of

phenol red, creatinine, inulin and urea clearances. It was inferred from these measurements that no marked or persistent decrease of renal blood flow had occurred. To confirm this view, determinations of renal blood flow and of mean femoral arterial pressure were made in uninephrectomized dogs with single explanted kidneys before and after compression of the renal artery by a metal clamp.

Hypertension and renal ischemia occured after clamping in most of these. In five, however, renal blood flow and function were only temporarily reduced after clamping, and returned to normal levels during the persistence of moderate hypertension.

These observations seem to exclude renal ischemia as the cause of renal hypertension. The most obvious circulatory change, other than ischemia, which might result from compression of an artery is reduction of pulse pressure distal to the clamp. The demonstration by Kohlstaedt and Page that renin is liberated from the isolated dog's kidney perfused with blood under constant mean arterial pressure, but with reduced pulse pressure, suggests, as do the observations here reported, that reduction of renal arterial pulse pressure rather than renal ischemia is the factor which maintains the arterial pressure of experimental hypertension.

Effects of desoxycorticosterone on fluid metabolism in the chronic hypophysectomized rat. E. L. Corey and S. W. Britton. *Physiological Laboratory, University of Virginia Medical School, University.*

As previously shown in this laboratory, hypophysectomized rats show a marked polyuria and polydipsia immediately after operation. A diminution in the diabetic condition is observed in 2 to 4 days, however, and at the end of 8 to 10 days after hypophysectomy a chronic but slight diabetes sets in and may persist until death. The present study, including data derived from over 250 individual tests, is particularly concerned with this phase of chronic, mild diabetes observed in hypophysectomized rats.

Desoxycorticosterone acetate injected into such animals (2 mgm. every 2 hrs. for 12 hrs.) resulted in a return of the severe diabetic condition observed initially, with marked hypochloruria. Thus, a series of 12 hypophysectomized uninjected rats observed from the 12th to the 32nd postoperative day showed a water intake of 2.7 cc., urine output of 2.1 cc., and chloride excretion of 4.44 mg., all determined on the basis of 100 grams body weight. The same animals in alternate tests with desoxycorticosterone showed values for water, urine and chloride of 8.2, 4.9 and 1.37 respectively. Thirty-four normal untreated rats showed respective values of 1.7, 1.5 and 4.47. Massive doses of desoxycorticosterone did not augment the diabetic, hypochloruric response.

In such series studied for extended periods it was observed that the effects of desoxycorticosterone were progressively diminished with repeated trials. An increased resistance or tolerance to desoxycorticosterone action (anti-hormone effect?) is therefore indicated. Thus, hypophysectomized rats tested with desoxycorticosterone from 32 to 64 days after operation showed water, urine and chloride levels of 4.3 cc., 2.5 cc. and 1.54 mgm. Alternate tests without injection yielded respective values of 2.2, 1.9 and 4.50.

The effects of desoxycorticosterone described above were completely antagonized by post-pituitary extracts. The vehicles used—sesame oil,

propylene glycol were apparently inactive as regards the diabetogenic effect.

Fundamental differences in the reactivity of autonomic and cerebrospinal nervous systems. R. CORTELL (by invitation) and E. GELLHORN. *Department of Physiology, College of Medicine, University of Illinois, Chicago.*[1]

Whereas somatic movements elicited by direct stimulation of the hypothalamus and the spinal cord, and by reflexes centered in the brain stem decline under the influence of anoxia, it is found that autonomic centers at these levels show an increased excitability during anoxia. As indicators of autonomic reactivity the nictitating membrane (N.M.), pilomotor, or blood pressure responses were studied during hypothalamic, medullary, and spinal cord stimulation.

The threshold for the pilomotor response resulting from either medullary or spinal cord stimulation is lowered during short periods of inhalation of 4.5–6.0 per cent oxygen. Similar concentrations of oxygen cause an increase in the height of the contraction of the N.M. elicited by hypothalamic stimulation in both normal cats and cats deprived of their "buffer nerves." The blood pressure rise resulting from hypothalamic stimulation in normal cats is increased during low oxygen inhalation. Unlike the N. M. response, however, the blood pressure response is diminished during anoxia, if the carotid sinus nerves and the vagi have been sectioned. The blood pressure rise resulting from medullary stimulation in the region of the vasomotor center likewise increases during anoxia. Furthermore, in some cases, a depressor response elicited by medullary stimulation may become a greater depressor response; in other cases, it may be converted to a pressor response.

The depression of somatic activity at the same time that autonomic activity increases under conditions of anoxia is an expression of fundamental differences between autonomic and cerebrospinal centers. Since similar differences are found under conditions of hypercapnia and hypoglycemia it is assumed that these characteristics of autonomic centers make adjustment reactions to alterations of homeostasis possible.

Determination of cardiac output in man by the direct Fick method and the ballistocardiograph.[2] ANDRÉ COURNAND and HILMERT A. RANGES (introduced by Homer W. Smith). *Departments of Medicine of the College of Physicians and Surgeons, Columbia University, and New York University College of Medicine, and the Third (New York University) Medical Division of Bellevue Hospital, New York City.*

A method of catheterization of the right auricle in man has been developed which permits the collection of mixed venous blood and the measurement of cardiac output by the direct Fick principle (O_2 and CO_2). A special 10-gauge Lindemann needle is introduced into the median basilic vein of either arm, and a no. 7, or no. 8, silk, radiopaque ureteral catheter, continuously but slowly perfused with saline, is introduced through the needle and slowly threaded up the vein. Catheterization is carried out on a horizontal fluoroscope table in order to guide the catheter to the desired

[1] Aided by a grant from the John and Mary R. Markle Foundation and W.P.A. Project 30278.
[2] Aided by a grant from the Commonwealth Fund.

position. When the tip of the catheter is determined to be in the right auricle, blood is collected by disconnecting the saline reservoir and attaching to the catheter a 3-way stopcock connected with two syringes, one filled with saline and one containing oil. Blood is first drawn into the saline syringe, then into the oiled syringe. Ten to 15 cc. of blood can be collected within 25 seconds with the slightest pressure.

Simultaneous femoral arterial blood is taken and expired air collected. Sampling of mixed venous blood and arterial blood, and collection of expired air are carried out with the patient lying on the ballistocardiograph. Ballistocardiograms are recorded just before and after sampling. There is no pain involved in passing the catheter, the pulse rate does not change before or during the procedure and the stroke volume as determined by the ballistocardiograph before, during and after the blood sampling and collection of expired air remains unchanged. The results do not appear to be affected by psychic disturbances.

Right heart catheterization has been performed 11 times and 4 successful comparisons of cardiac output by direct Fick and ballistocardiograph have been made. The observations are being continued and a detailed report will be made.

Experimental macrocytic hyperchromic anemia. LATHAN A. CRANDALL, JR., C. OSVILLE FINNE, JR. (by invitation) and PAUL W. SMITH. *Department of Physiology, University of Tennessee College of Medicine, Memphis.*

In the course of other investigations on dogs with "internal" biliary fistulae (anastomosis of gall bladder to right renal pelvis with ligation of common duct) it was observed that the anemia developing spontaneously in these animals is of the macrocytic hyperchromic type, similar in blood picture to pernicious anemia and sprue in man.

We have now followed the progress of 12 such dogs, only one of which has failed to develop a "primary" type of anemia within 4 months after operation. Deficiencies in vitamins A, D, and K also are apparent. Appropriate treatment of these vitamin deficiencies has not altered the course of the anemia.

In eleven of the bile fistula dogs the mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCHb) have shown increases exceeding 2 standard deviations from the mean for normal animals and often have exceeded 3 S. D. Parenteral administration of iron, copper, alpha-tocopherol, thiamin, riboflavin, nicotinic acid, and pyridoxin has had no effect on the development of the anemia. The anemia has occurred when 4 grams of yeast have been added to the daily diet.

The anemic animals respond to the parenteral administration of purified liver extract (Lilly, 15 units per cc.) by reticulocytosis, return of MCV and MCHb to within normal limits, and by increases in red cell count and hemoglobin concentration.

These observations are believed to indicate that absence of bile from the digestive tract interferes with the absorption of the erythrocyte maturation factor.

Muscle and cardiac electrolyte in potassium poisoning. J. M. CRISMON,[1] C. CRISMON, M. CALABRESI and D. C. DARROW (introduced by Victor

[1] On leave of absence from Department of Physiology, Stanford University.

E. Hall). *Department of Pediatrics, Yale University School of Medicine, New Haven, Conn.*

Potassium poisoning as indicated by A.V. block in an electrocardiogram was produced by intraperitoneal injection of 0.5 molar KCl in pentabarbitalized cats. Analyses of serum, muscle and heart were carried out for the principal electrolytes. In some experiments the concentration of serum electrolyte was changed by simultaneous injection of hypertonic NaCl-$NaHCO_3$ and in others, by removal of extracellular electrolyte by the intraperitoneal injection and subsequent removal of 5 per cent glucose.

A.V. heart block develops when the concentration of potassium in the serum rises to 8 to 12 mM per l. Potassium usually increases considerably in the heart and somewhat less in the muscle but these tissue changes do not occur in all experiments. When hypertonic NaCl-$NaHCO_3$ is injected along with 0.5 molar KCl, the rise in cardiac K is greater than when KCl alone is injected. A.V. block occurs, however, at the same serum level. When extracellular electrolyte has been depleted, A.V. block occurs at a somewhat higher serum level than in most of the other experiments but cardiac potassium is essentially normal. In one experiment the simultaneous injection of 5 per cent glucose with 0.5 molar KCl produced elevation of serum potassium to 9 mM without electrocardiographic signs of K poisoning although cardiac potassium reached a high level.

The results confirm previous observations on the relation of the concentration of potassium in serum to heart block; they show that cardiac potassium usually rises under these circumstances but A.V. block may occur when cardiac potassium is normal when expressed per unit of fat free solids or per unit of intracellular water.

Reflex modification of respiration by intestinal distention. Robert T. Crowley (introduced by Charles G. Johnston). *Department of Surgery, Wayne University College of Medicine, Detroit, Mich.*

Thirty dogs were used in the course of this experiment. Under complete anesthesia produced by injections of pentobarbital, the spinal cord was exposed at the level of the first dorsal vertebra, the vagus nerves isolated, the right carotid artery cannulated, a pleural cannula inserted in the right chest, and a distending tube placed in the small intestine 24 inches from the pylorus. Continuous respiratory and blood pressure tracings were recorded simultaneously on the kymograph. Periodically the tube in the intestine was inflated to produce intraluminal pressures varying from 50 to 150 mm. Hg.

Distention of the small intestine produced marked respiratory changes in all of the experimental animals employed. The respiratory effect consisted of a short period of apnea or irregular breathing followed immediately by a marked increase in volume and usually in rate. The rapidity and the magnitude of the respiratory effect were directly proportional to the speed with which the distension of the gut was produced and also directly proportional to the amount of distention, 100 mm. Hg giving a more pronounced effect than 50 mm. Hg.

Bilateral section of the vagi did not abolish the respiratory effect, but transection of the spinal cord at the level of the first dorsal vertebra or isolation of the distended gut segment from all mesenteric connections eliminated it completely.

Anesthesia depressed the respiratory effect in direct proportion to the depth of the narcosis induced. Cocainization of the mesentery attached to the distended segment with a 1 per cent solution depressed the respiratory effect but frequently did not abolish it after twenty minutes. Atropine (5 m./kg.) and yohimbin (6 mg./kg.) intravenously did not abolish the respiratory effect.

Concomitant alterations in systolic blood pressure occurred with the appearance of the respiratory changes, the most usual effect being an immediate elevation upon distension and fall after release.

The circulation in traumatic shock. M. L. CULLEN (by invitation), A. E. SCHECTER (by invitation) and N. E. FREEMAN. *Harrison Department of Surgical Research, School of Medicine, University of Pennsylvania, Philadelphia.*

Traumatic shock was produced in nine out of eleven dogs and in five partially sympathectomized dogs. Local fluid loss was excessive in all cases. The earliest sign of shock was a marked reduction of peripheral blood flow. Hemoconcentration was not a significant finding.

Shock was produced in fourteen out of sixteen dogs with more severe trauma, but with restriction of local fluid loss. The earliest sign of shock was again a marked reduction of peripheral blood flow. Hemoconcentration occurred at a later time.

Blood volume determinations by the carbon monoxide method showed a substantial reduction of blood volume after trauma, and this reduction was only partially accounted for by local fluid loss.

Post mortem examinations suggest that at least part of the "lost" blood volume is to be found in the lumen and mucosa of the upper intestinal tract.

Membrane resting and action potentials of the squid giant axon. HOWARD J. CURTIS and KENNETH S. COLE. *Department of Physiology, Johns Hopkins University School of Medicine, Baltimore, Md., and Department of Physiology, Columbia University, New York City.*

The action and resting potentials from the membrane of the squid giant axon have been measured between one electrode inside and another electrode outside the axon. The membrane is pierced at one end of the axon by a capillary glass needle filled with isotonic KCl which is then pushed along the axis of the fiber for a distance of 15 mm. Potentials are recorded between large Ag-AgCl electrodes leading to this capillary and to one on the outside of the fiber directly opposite the tip of the needle. These impaled axons often remain excitable for many hours, and experiments were discarded unless excitability was maintained for at least an hour after inserting the needle.

The resting potential of these axons varied from 46 to 59 millivolts and averaged 51 millivolts. Liquid junction potentials undoubtedly cause a slight error in these measurements. Action potentials recorded in this way were somewhat diphasic, and tended to be oscillatory, with the first positive phase about 15 per cent of the spike height. The maximum negative variation from the resting potential (spike height) varied from 77 to 168 millivolts and averaged 108 millivolts. Thus during the passage of an impulse the membrane potential is momentarily reversed in sign

so that the outside may be as much as 110 millivolts negative with respect to the inside. The fact that the action potential is larger than the resting potential can be qualitatively explained by the observed membrane inductance.

The resting potential was measured as a function of the potassium ion concentration of the fluid surrounding the axon. It was found that in the region of the normal potassium concentration there was a relatively small change of potential with concentration, but at higher concentrations the potential fell sharply, reached zero at about 20 times normal concentration, and was about 15 millivolts negative at 40 times (isotonic KCl). The spike height was very sensitive to increases in potassium concentration, and was reduced to only a few millivolts by a concentration which left the resting potential practically unchanged.

Changes in the rhesus uterus during labor. D. N. DANFORTH and R. J. GRAHAM (introduced by A. C. Ivy). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Frozen sagittal sections have been made of 30 Macacus Rhesus monkeys at various stages of late pregnancy, labor and the puerperium. Measurements made from the specimens themselves and from direct outline tracings reveal the following: During the first stage of labor, retraction appears to be more or less limited to the isthmus uteri, though the percent of retraction of the isthmus and corpus in proportion to the internal circumference contributed by each appears to be about the same. The cervical lips are retracted to and above the level of the symphysis pubis. During the second stage of labor retraction occurs at an essentially equal rate in corpus, isthmus and cervix. "Thinning of the lower uterine segment," commonly observed at Caesarian section in the human being, appears to be due only to the contrast between the fully retracted corpus, which was considerably better developed to start with, and the less hypertrophied, but nevertheless fully retracted structures which have been pulled upward from below. Deep anesthesia may also be a factor. In the third stage of labor retraction is most marked in the corpus uteri. After the completion of labor there is almost instant return of the uterus to the general form manifest prior to pregnancy.

Autonomic concomitants of changes in the electroencephalographic "spectrum." C. W. DARROW and M. L. PHILLIPS (by invitation). *Institute for Juvenile Research and Department of Physiology, University of Illinois, Chicago.*

By a method providing continuous graphic analysis of the frequency of the human electroencephalogram along with simultaneous systolic blood pressure, palmar galvanic (sweating) and respiratory changes certain patterns of response are shown to be of significance in a relatively large proportion of individuals. Sympathetic activity as indicated by simultaneous rises in systolic blood pressure and palmar sweating are shown to be attended or immediately preceded in a large proportion of instances by one or more of the following: 1, increase in "beta" activity; 2, blocking of "alpha," or 3, alpha out of phase in the two hemispheres. The increase of alpha appears associated with decrease of the prevailing sympathetic tone. Parasympathetic activity as inferred from a fall in blood pressure following stimulation is attended by augumentation of certain of the slower

frequencies in those individuals manifesting these frequencies. In such individuals, inhibition of parasympathetic tonus as indicated by a rise in blood pressure without concomitant sympathetic palmar sweating is attended by a decrease in slow potentials. Certain exceptions to these interrelationships are specified in the attempt to define the limits within which it is justifiable to derive an autonomic interpretation of the electroencephalogram.

Effect on pulse rate of peripheral arterial occlusion and release.[1] D. V. DAUBER, H. WEINBERG and M. LANDOWNE[2] (introduced by L. N. Katz). *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.*

The effect on pulse rate of applying and releasing occluding cuffs to the lower extremities was observed in 27 normal and in 4 subjects with thromboangiitis obliterans. Fifty-seven tests, each involving two or more occlusions, were performed with the subjects horizontal. Pulse rates were measured from electrocardiographic records. A rapid interruption of the arterial inflow was effected by connecting wide cuffs placed around the proximal thighs to a pressure tank at over 200 mm. Hg. The immediate return of flow was assured by disconnecting the pressure tank and opening the cuff tube.

In about half of the cases, application of the occluding pressure was followed by a slowing in pulse rate of 4–20 beats per min. (average 9), developing 1–3 sec. after occlusion and lasting 5–16 sec. During the period of occlusion, no significant changes in pulse rate occurred. On release of the occluding pressure, a pulse acceleration of 15 to 30 beats per minute was noted in 97 per cent of the cases, lasting $\frac{1}{6}$ to 3 minutes. This acceleration commenced 1 to 3 sec. after release, reaching its maximum in four to five seconds. The extent and duration of the pulse acceleration varied directly with the length of prior occlusion. No rise in pulse rate occurred on release of arterial occlusion in the four patients with thromboangiitis obliterans.

A fall in arterial blood pressure was observed to follow release of the arterial occlusion; and in 7 cases studied by direct needle puncture of the brachial artery with a Hamilton manometer, this blood pressure fall was observed to be instantaneous and the pulse contour changed as expected with a lowering of peripheral resistance. The blood pressure dropped from 12 to 35 mm. systolic and from 14 to 27 mm. diastolic. An instantaneous, transient rise in blood pressure occurred on application of the occlusion in the five cases studied with the same technic.

It is concluded from the time at which the blood pressure and pulse rate change occurred that the pulse acceleration on release of peripheral arterial occlusion is a reflex response to the drop in blood pressure, and that the latter results from the lowering of the peripheral resistance by reopening the leg vascular channels. The failure of patients with narrowing of the peripheral vessels to develop a similar pulse acceleration would favor this interpretation.

The reaction of living bone to various metals and alloys. H. A. DAVENPORT and R. T. BOTHE (introduced by W. F. Windle). *Department of Anatomy, Northwestern University Medical School, Chicago, Ill.*

[1] Aided by the A. D. Nast Fund for Cardiac Research.
[2] Emanuel Libman Fellow.

A sequel to work reported previously (Surg., Gynec. and Obstet. **71**: 598, 1940), consisted of implanting aseptically into the femurs of adult cats, 2 mm. pegs of the following metals: Al, Ag, Be, Cd, Co, Cr, Cu, Mn, Mo, Ni, Pb, Ti, and W; and the alloys: Au-Pt, Mn-Ti, Cr-Mn, Fe-Ti, stainless steel and vitallium. The implants were driven into holes drilled through the cortex of the bone and into the medulla, so that the metal came in contact with cortex, medulla, and periosteum. Two implants usually of dissimilar metals were made in each femur. Reactions of the bone were characteristic for each metal. Readings of differences in electrical potential between couples of dissimilar metals as related to the reactions gave no support to the idea sponsored by some surgeons that electrolysis is the cause of unfavorable bone reaction to metallic prostheses. Ti, Pb, Cr, Al, vitallium and stainless steel were tolerated well. Cu was toxic, as shown by bone recession from the implant; Mn stimulated callus formation with covering of the implant by new bone formation, and Co became enclosed in a fibrous tissue capsule within an eroded area. It is concluded that living bone reacts to adjacent metal in the manner conditioned by the solubility of the metal in body fluids plus the effect of the metallic salts so formed. Toxicity of salts of metals, such as lead and chromium, is not necessarily a criterion when the metals themselves are insoluble.

The inhibition of carbonic anhydrase and gastric acid secretion by sulfanilamide. HORACE W. DAVENPORT (introduced by C. N. H. Long). *Department of Physiological Chemistry, Yale University Medical School, New Haven, Conn.*

Sulfanilamide is a powerful inhibitor of carbonic anhydrase when the enzyme is in dilute solution, and the relation between the enzyme and the inhibitor follows the mass action law. However when the concentration of carbonic anhydrase is increased the inhibition by sulfanilamide becomes weaker. At enzyme concentrations approaching those in the parietal and red blood cells very high concentrations of sulfanilamide of the order of 20–100 mgm. per cent are necessary to produce effective inhibition of the enzyme. Consequently it can be predicted that only high concentrations of sulfanilamide can produce a significant decrease in the ability of carbonic anhydrase to catalyse the hydration and dehydration of carbon dioxide in the body. This prediction is confirmed by clinical experience and by the direct observations of Roughton and his colleagues (unpublished data).

Sulfanilamide has a small but definite inhibitory effect on the secretion of acid by the gastric mucosa of dogs. At the plasma sulfanilamide concentration of 25 mg. per cent the inhibition of the rate of acid secretion is from 20 to 40 per cent. Below 10 mgm. per cent the inhibition is negligible. These observations agree with the order of magnitude of inhibition to be expected, and they support the theory that carbonic anhydrase is an integral part of the acid secreting mechanism.

A new microrespirometer for nerve. P. W. DAVIES and FRANK BRINK, JR. (introduced by D. W. Bronk). *Department of Physiology and Biophysics, Cornell University Medical College, New York City.*

Heyrovsky's electrode method (O. H. Müller, Cold Spring Harbor

Symposia **7**: 59, 1939) has been used in a microrespiration chamber which is suitable for measuring respiration of nerve. A 27 micron platinum-iridium wire is sealed into the wall of a small glass tube (4 cm. x 2 mm.), the tip of the wire projecting into the lumen. The tube is filled with a solution and the electrode is made 0.9 volt negative with respect to a calomel half-cell which communicates with the chamber. The current which then flows is proportional to the oxygen concentration of the fluid. A respiring nerve placed in the chamber reduces the oxygen concentration of the solution, causing a progressive decrease in this electrode current. After a ten minute diffusion transient the current decreases at a constant rate which is a measure of the nerve's respiration. Our apparatus will detect a consumption of approximately 10^{-3} cu.mm. oxygen and can be used to follow minute by minute the respiration of 10 mg. of peripheral nerve.

To calibrate the cell a larger auxiliary wire is placed in the chamber and made cathodal. The constant current through this calibrating wire electrolyzes oxygen from the solution ($2H^+ + O_2 + 2\epsilon \rightarrow H_2O_2$) at a definite rate. The resulting change of oxygen concentration is measured by the small measuring electrode as previously described. The rate of change of current in this measuring electrode is found to be approximately proportional to the rate of removal of oxygen from the solution. Complicated diffusion corrections are thus avoided.

This method has three advantages over the volumetric micromethods. The system is quite insensitive to temperature changes, making accurate temperature control unnecessary. Secondly, it is easy to change the solution bathing the nerve by flowing the new solution in through a capillary at the end of the tube. Thus successive respiration measurements on the same nerve can be made quickly. A third advantage lies in the use of electrical measurements which can be made as accurate as desired.

Values of nerve respiration obtained by this method are comparable with those measured volumetrically.

Factors affecting the electroencephalographic changes induced by hyperventilation. H. DAVIS and W. MCL. WALLACE (by invitation). *Departments of Physiology and Biochemistry, Harvard Medical School, Boston, Mass.*

EEGs were recorded during and after 52 standardized hyperventilation tests on 6 healthy male subjects. Blood from a finger pad, taken just before and just after hyperventilation, was analyzed for total CO_2, pH (serum) and blood sugar. Breathing was timed by metronome at 15 breaths per min. for 3 min. and the depth adjusted by instructing the subject according to the excursion of the drum of a Roth-Benedict metabolism apparatus to 20 cc. (1 mm. on scale) per pound of body weight. As arbitrary measures of the modification of the EEG, in addition to the usual visual inspection, we counted the number of waves above an arbitrary size (never reached in normal control records) passing an electrical filter broadly tuned to 5 cycles, and we also measured a modified "delta index" (Hoagland).

Repeated tests on the same subject on the same day at intervals of 20 to 40 min. produced closely similar changes in total CO_2, in pH and in

the EEG. CO_2 tension fell 18 to 20 mm. Hg, and pH increased by 0.15 to 0.20 units. The standardization of breathing seemed adequate to produce a standard and considerable respiratory alkalosis.

Metabolic alkalosis (pH up to 7.53 before hyperventilation) and acidosis (pH down to 7.29) produced by oral ingestion of $NaHCO_3$ and NH_4Cl respectively caused no significant change in the effect of hyperventilation on the EEG.

Hyperventilation with oxygen produced less alteration than with air, even when the oxygen absorbed from the air in the metabolism apparatus was continuously replaced.

Low blood sugar favors the appearance of delta waves, and a high sugar level (over 100 mgm. per 100 cc.) strongly tends to prevent their appearance. At a given blood-sugar level, there are great individual differences in the amount of slow-wave and delta activity that will appear, and some subjects may vary considerably in their responses from one day to another, but the effect of low blood sugar is so important that blood sugar must be controlled if hyperventilation is to be standardized as a test for stability of the EEG.

The production of experimental polycythemia in dogs and rabbits by the daily administration of ephedrine. JOHN E. DAVIS (introduced by F. J. M. Sichel). *Department of Physiological Chemistry and Pharmacology, University of Vermont College of Medicine, Burlington.*

We have previously shown (1940) that choline and other vasodilator drugs are effective in depressing the experimental polycythemias produced in dogs by cobalt or by exposure to low atmospheric pressures. To explain this action, we assume that these dilator drugs improve the blood flow to bone marrow, thus diminishing the local anoxia and removing the stimulus to polycythemia.

If this explanation is correct, it would seem that drugs which might constrict marrow arterioles should have the opposite effect; i.e., should increase the rate of red blood cell formation.

Ephedrine sulphate has been administered orally to 4 normal dogs in daily doses ranging from 2.5 to 5 mgm. per kilogram. After about ten days on this routine, each dog exhibited an increase of about one million in his basal erythrocyte number. The increases occurred gradually, and were not accompanied by increased leukocyte counts. The percentage of reticulocytes was almost doubled. The red cell counts dropped to normal within 7 to 10 days after cessation of ephedrine.

In similar experiments on rabbits, two normal and two splenectomized animals have been given 45 mgm. of ephedrine sulfate daily by subcutaneous injection. Within 6 to 12 days each rabbit showed an increased number of erythrocytes (increase about 1 million). Reticulocyte percentages increased from about 1.5 to 4.0 (average). Hemoglobin percentages were also increased.

These results can be explained by assuming that ephedrine reduces the blood flow to bone marrow, thus reducing oxygen supply, and stimulating erythropoiesis.

Effect on the electroencephalogram of alterations of blood sugar level. P. A. DAVIS (introduced by H. Davis). *Department of Physiology, Harvard Medical School, Boston, Mass.*

Thirty fasting college students were each given insulin intravenously (U-40 or 0.00056 cc. per lb. body weight). EEGs and EKGs were then recorded continuously for over an hour, during which 7 blood samples were taken.

The lowest blood-sugar levels occurred between 25 and 30 minutes post-insulin, but levels reached (53 to 75 mgm. per 100 cc.) were independent of the original levels (80 to 113 mgm. per 100 cc.). As the blood sugar falls, the EEG changes appear, but the sequence of these changes is independent of the rate of fall or subsequent rise.

The sequence of EEG changes is as follows: In 15 to 20 minutes a reduction of alpha and the appearance of 8 per second waves occur. In 20 to 25 minutes, 4 to 8 per second activity dominates. By 30 minutes delta activity and also very slow sinusoidal waves, 2 to 5 seconds long, appear. The latter may appear simultaneously with the usual delta, the 4 to 8 per second or alpha activity, and may recur until the 45th minute. At 40 minutes low voltage 16 to 20 per second waves gradually come in, replacing the 4 to 8 per second activity until the alpha returns and finally dominates the picture and the EEG becomes stable again.

In 4 cases of EEGs with inherent slow dysrhythmia, the slow activity was more clearly organized and stationary when the blood sugar was at the lowest level.

The sympathetic reaction, indicated by acceleration of the pulse, occurs between 25 and 30 minutes after the EEG changes have begun. Therefore it is the fall in blood sugar and not the sympathetic reaction which causes the instability of the EEG. This instability continues approximately 10 minutes after the pulse has resumed its previous rate. Ordinarily the EEG remained unstable, and the 4 unusual cases referred to maintained organized dysrhythmic activity, for 5 to 10 minutes after the rising blood sugar had passed the level at which the EEG first changed when the blood sugar was falling.

Kinetics of lung ventilation with special reference to the use of helium.

ROBERT B. DEAN (by invitation) and MAURICE B. VISSCHER. *Department of Physiology, University of Minnesota, Minneapolis.*

Helium requires about 10 per cent more pressure than N_2 for equal velocities of streamline flow. However, a mixture of 80 per cent He, 20 per cent O_2 can flow approximately three times as fast as air before its motion becomes turbulent; consequently the pressure necessary to maintain a turbulent flow of air is greater than the pressure necessary to maintain the same velocity in a He-O_2 mixture. Calculations show that turbulence in the normal dog lung occurs only in the trachea and larger bronchioles.

Although diffusion of He is nearly 4 times as rapid as N_2, a model containing contorted passages smaller than the smallest in the lung showed essentially equal flows of air and He; indicating that diffusion can not be a major factor controlling the ventilating pressure of the lung.

We have studied dog lungs ventilated by a simple pump connected to the trachea using a glass spoon manometer to record pressures. Pressures were plotted against volumes displaced by the pump to produce curves for a ventilation cycle similar to those obtained mechanically by Bayliss and Robertson (Quart. J. Exper. Physiol. **29**: 27, 1939).

The resistance to ventilation can be separated into a viscous component, which includes any turbulence as well as tissue viscosity; and an elastic component which is entirely due to the tissues. The elastic but not the viscous component is reduced by opening the chest. Substitution of He-O_2 mixtures for air produces no change in the viscous resistance, and slowing the pump from 20 to 6 cycles per minute produces only a small decrease in viscous resistance. Some of the tissue elasticity appears to be due to the fact that rapid inflation causes an increase in pressure which falls slowly if the volume is maintained and is considerably lowered by a small reduction in volume.

The interaction of cortical potentials produced by stimulation of the thalamus. E. W. Dempsey and R. S. Morison. *Departments of Physiology and Anatomy, Harvard Medical School, Boston, Mass.*

Stimulation of peripheral nerves in cats under nembutal anesthesia is followed by short latency responses which are sharply localized in the sensorimotor cortex. These responses decline in magnitude as the stimulus frequency is increased, and have an electrical sign unrelated to that of the prevailing 8–12/ sec. spontaneous activity. Similar responses are produced by stimulation of points within the cortical relay nuclei in the lateral thalamic mass.

Stimulation of the medial thalamic nuclei gives rise to widespread, long latency responses in the frontal cortex. These effects increase rapidly in size (recruit) on repetitive stimulation at frequencies up to 10/ sec., after which alternation or disappearance occurs. The polarity of these potentials is the same as that of the 8–12/ sec. spontaneous activity.

The effects of peripheral nerve or lateral thalamic stimulation occur on a quiet background or may be superimposed upon both the spontaneous activity and the recruited response. The recruited response, on the other hand, is blocked by simultaneously occurring spontaneous activity.

Since spontaneous activity does not render the thalamic relay nuclei refractory, it follows that the spontaneous activity cannot be due to reverberating circuits between the cortex and relay nuclei. On the other hand, since there is refractoriness to the recruited response during spontaneous activity, it is concluded that the medial nuclei are an integral part of a mechanism controlling the 8–12/ sec. rhythm. This conclusion is reinforced by the observation that destruction of the medial nuclei permanently abolishes or greatly reduces spontaneous activity (Morison and Dempsey, Proc. Am. Assoc. Anat., 1941).

Some observations of the effects of nitrous oxide upon the electroencephalogram in man. A. J. Derbyshire, F. J. Murphy (by invitation), K. E. Corrigan (by invitation) and L. Lobdell (by invitation). *Department of Anatomy, Wayne University College of Medicine and Harper Hospital, Detroit, Mich.*

These are preliminary experiments in an attempt to evaluate the degree of oxygen lack coincident with the administration of various nitrous oxide—oxygen mixtures. A correlation between known anaesthetic mixtures, the EEG and subjective reports was made which may be of clinical interest.

On two abnormal patients mixtures of 50-50 nitrous oxide—oxygen to pure nitrous oxide were used, while on eight normal subjects mixtures of

50-50 to 95-5 and the corresponding nitrogen—oxygen mixtures were employed. Standard EEG technique with multiple monopolar leads was applied. Subjective reports during and after each experiment were recorded on phonograph records.

With increasing percentages of nitrous oxide the following changes were observed: 1. With mixtures up to 70 nitrous oxide—30 oxygen the number of alpha waves per 30 seconds was decreased, first in the motor area and subsequently in the occipital area. 2. When mixtures of 70 nitrous oxide—30 oxygen to 90 nitrous oxide—10 oxygen were given frequencies of 4 to 8 per second appeared in most subjects and were dominant in the motor areas. 3. With pure nitrous oxide two possible patterns appeared. *a.* A 2.5 per second pattern of 150 uV amplitude occurred if the induction was rapid. *b.* If the patient had a three minute induction with a 50-50 to 70-30 mixture followed by pure nitrous oxide, an irregular delta pattern of about 40 uV was produced. Both patterns (*a* and *b*) started about the time that cyanosis of the nail beds appeared. Wide individual variations in EEG patterns were encountered for each gas mixture but so far each subject has followed in his particular way the above described plan.

In contrast, mixtures of nitrogen and oxygen given to normal subjects produced no change until less than 10 per cent oxygen was used. Normal subjects have been exposed to the very low oxygen mixtures only long enough to effect an increase in voltage and the appearance of slow, 8 to 4 per second rhythms.

Reabsorption of the nutritionally essential amino acids by the kidney tubules.[1] J. R. Doty (by invitation) and A. G. Eaton. *Department of Physiology, Louisiana State University School of Medicine, New Orleans, La.*

A previous report (Proc. Soc. Exper. Biol. and Med., in press) has indicated that the kidney tubules can reabsorb certain amino acids at a very rapid rate. The present study was designed to measure the rates of reabsorption of those amino acids generally considered to be essential constituents of the diet. This study would seem to be the first step in the investigation of the mechanism of tubular reabsorption of the amino acids, as well as an additional step in the attempt to correlate intestinal absorption and tubular reabsorption.

A solution of sodium ferrocyanide and the amino acid under investigation was administered to dogs by intraperitoneal injection. The volume of glomerular filtrate was calculated from the plasma level of ferrocyanide and the amount of ferrocyanide excreted. The amount of amino acid in the glomerular filtrate was then obtained by determination of its concentration in the plasma. Analysis of the urine for the period gave by difference the amount of the amino acid reabsorbed. The rates of reabsorption will, of course, be higher than indicated if there is any marked "tubular excretion" of amino acids. It seems unlikely, however, that tubular excretion is significant at the plasma concentrations attained in these experiments.

Several of the widely used protein precipitants were found to be unsuitable for the preparation of the protein free plasma filtrates employed in this

[1] Aided by a grant from the Committee on Scientific Research of the American Medical Association.

work. The use of a freshly prepared solution of trichloracetic acid gave best results on recovery trials. The amino acid content of the plasma was determined by the manometric method of Van Slyke as well as by certain colorimetric methods in individual instances.

The results indicate a considerable variation in the ability of the tubules to reabsorb different amino acids.

An analysis, by hydraulic models, of the factors operating to produce the typical ballistocardiogram. PHILIP DOW and W. F. HAMILTON. *Department of Physiology and Pharmacology, University of Georgia School of Medicine, Augusta.*

An attempt has been made to evaluate the many factors which operate to produce the characteristic sequence of recoils responsible for the ballistocardiogram. Hydraulic models have been designed to simulate the cardiac recoil, the aortic recoil, and the oscillations in the aorta associated with its standing waves. The behavior of the forces in these models has been recorded on the vertical ballistocardiograph described in a companion report.

In all experiments water was forced from a bottle through a large opening by inflation with air of a cloth-enclosed rubber bag in the bottle. The rate and extent of inflation were reproducibly controlled by a stopcock, by a graded series of side chambers in the air line, and by the level of the air pressure.

The recoil of a simple jet is a monophasic curve which becomes diphasic at the very end of ejection only if the rigid outlet tube is lengthened. If the ejection is made into a vertical "aorta" of rubber tubing, the ballistogram becomes oscillatory, in tune with the standing waves in the tube. If this "aorta" is now arched, the frequency of the ballistogram waves is doubled, with an asymmetry which depends upon the relative lengths of the two arms of the arch.

The form and magnitude of the ballistographic waves have been checked against simultaneously recorded pulses from various parts of the rubber aorta. They have also been compared with the total output, the rate of ejection, and the initial acceleration as recorded by a sensitive tambour from a tight jar containing the whole model. The form and frequency of the waves seem to be definitely associated with the fundamental and first octave standing waves in the aorta of the model. The size of the initial deflection depends not at all upon the total stroke output, but rather upon the acceleration, or the rate at which the velocity of ejection is built up.

Grateful acknowledgment for aid in this research is made to the Josiah Macy, Jr. Foundation.

Lipocaic and ketonemia in pancreatic diabetes. LESTER R. DRAGSTEDT, DWIGHT E. CLARK (by invitation), ORMAND C. JULIAN (by invitation), J. GARROTT ALLEN (by invitation) and CORNELIUS W. VERMEULEN (by invitation). *Department of Surgery, The University of Chicago, Chicago, Ill.*

The relation of lipocaic to fat metabolism is indicated by the occurrence of marked hypolipemia and severe fatty infiltration of the liver in depancreatized dogs with lipocaic deficiency. The administration of lipocaic to such animals elevates the blood lipids to normal and clears the liver of fat.

The accompanying marked increase in glucose excretion and insulin requirement suggests that under the influence of lipocaic, fat in the liver may be converted into glucose. The present study was devised to determine the relation, if any, between lipocaic and ketogenesis. The concentration of ketone bodies in the blood of depancreatized dogs was determined under the following conditions: *a*, depancreatized dogs adequately controlled with both insulin and lipocaic; *b*, depancreatized dogs receiving adequate insulin but no lipocaic; *c*, depancreatized dogs receiving adequate lipocaic but inadequate insulin therapy; *d*, depancreatized dogs with fatty liver of lipocaic deficiency adequately controlled with insulin and then given curative doses of lipocaic.

In these various conditions, abnormal elevation of the blood ketones occurred only with inadequate administration of insulin, and no relation to lipocaic deficiency or overdosage was observed.

Further studies on the origin of glycoside emesis. MELVIN DRESBACH. *Harrison Department of Surgical Research and the Department of Physiology, School of Medicine, University of Pennsylvania, Philadelphia.* (Read by title.)

Recent studies directed to explain the mechanism of glycoside emesis made it increasingly obvious that further attempts to solve the problem by observing the actions of glycosides after denervation operations confined to the heart, or other individual viscera, would fail, as they have in the past (Am. J. Physiol. **126:** 480, 1939). The following operations were therefore performed, in separate stages, in the order designated: 1, dividing the spinal dorsal roots from C_3 to C_8 inclusive; 2, transection of the spinal cord at lever T_1 or T_2, with division of the remaining dorsal roots up to C_6; 3, mid-cervical, bilateral vago-sympathetic block by anesthesia, and later by surgical division. Eight dogs have been used thus far, four dying of complications before the third operation. Of the remaining four, one survived the cord transection and rhizotomy, carried upward to C_7 inclusive, and right, intra-thoracic vagotomy but lived only ten hours after mid-cervical left vagotomy. Two survived cord transection, rhizotomy to C_7 inclusive and bilateral vagal block (novocaine, 2 per cent; nupercaine, 1:200). The fourth withstood rhizotomy from C_3 to C_6, cord transection and division of dorsal roots remaining above; finally nupercaine block of the vagi and later double vagotomy, thus interrupting all known afferent nerve paths from the trunk. One cat was also operated, the rhizotomy going only to C_7 inclusive. Nausea and vomiting reactions (diaphragm contractions) were produced by strophanthidin in the first three dogs in this group and by one or more glycosides in two of them, after the final operation and within a month of the initial one. The fourth dog, with the most extensive denervation, reacted typically to strophanthidin, lanatosids, A, B and C, and apomorphine, within the one-month time limit arbitrarily set. Moreover, a significant increase in dosage in order to get emetic responses postoperatively was unnecessary. Positive results were likewise obtained in the single cat. It is plain that glycoside emetic responses can be induced in the dog (and doubtless the cat) independently of afferent innervation of all structures below the neck. The study continues.

Effects of hexylresorcinol and other agents on the absorption of sugars, chloride and sulfate from the alimentary tract. ROBERT L. DRIVER

(introduced by J. R. Murlin). *Department of Vital Economics, University of Rochester, Rochester, N. Y. and the Department of Anatomy and Physiology, University of Kentucky, Lexington.*

The absorption of sugar falls off with time as determined by the Cori technique, but it is doubtful if this method comes close enough to a normal physiological condition either to assume or to preclude a special mechanism for the absorption of sugars. Neither the lowering of surface tension nor the removal of calcium ions affect absorption, but in most cases absorption is decreased when the saccharide-splitting enzymes are inhibited. The inhibition of these enzymes can be effected by organic molecules containing specific hydrophobic and hydrophilic groups.

Chloride, in the presence of a polyvalent anion, leaves the gut against a considerable concentration gradient, but the data obtained refute any theory of differential permeability which is based essentially on diffusion. The action of hexylresorcinol strongly indicates that a special biological agent is responsible for the absorption of electrolytes as well as for other substances. It is suggested that this mechanism, as far as chloride is concerned, is at least partly mediated by carbonic anhydrase since hexylresorcinol inhibits both the action of the enzyme and the absorption of chloride.

The effect of exercise on ketone body metabolism. D. R. Drury and A. N. Wick (by invitation). *Department of Physiology, School of Medicine, University of Southern California, Los Angeles, and the Scripps Metabolic Clinic, La Jolla, Calif.*

Changes in ketone body concentration in the blood, and in output in the urine as a result of exercise were followed in the human subject. Ketosis was produced by having him take a constant ketogenic diet for several days. Urine excreted during the night (period of rest) always contained much more ketone bodies than that of the day (period of activity) except that when the subject remained in bed all day the output of the two periods became equal. When the activity of the day time period was increased by added exercise the urine output at this time became less, but the night period that followed this tended to increase above ordinary night periods.

The blood ketone body concentration became definitely lower as a result of a twenty minute period of moderate running but in the rest period following, its concentration came back to the pre-exercise level within an hour. After three hours of exercise the blood ketone concentration was not diminished.

These findings support the view that ketone bodies are used as source of energy in muscle activity and that the production of these substances by the liver is increased by exercise.

Studies on the electrocardiogram of the horse. H. H. Dukes and H. T. Batt (by invitation). *Department of Physiology, New York State Veterinary College, Cornell University, Ithaca, N. Y.*

We have made numerous electrocardiograms on horses by means of the string galvanometer. Most of the animals were patients in the large-animal clinic of this College, but many of them showed no evidence of heart disorder. In the earlier work leads from the pectoral (L.A.), dorsal (R.A.), and sternal (L.L.) regions were used but later limb leads were adopted.

With limb leads the P waves may be upward, inverted, diphasic, notched

or double. The R deflection is usually upward. The T wave is usually diphasic, but upward and inverted T waves are common.

In 17 records of lead II (mostly limb leads, a few chest leads) the following average intervals and ranges of variation were noted: P-R, 0.30 sec. (0.20–0.42 sec.); QRS, 0.12 sec. (0.08–0.17 sec.); QRS-T, 0.54 sec. (0.46–0.62 sec.).

Among the disturbances in rate or rhythm that we have observed in horses are the following: sinus arrhythmia, ventricular premature beats, various stages of heart-block (partial heart-block is not uncommon in the horse—Roos), atrial fibrillation under chloroform anesthesia, and naturally occurring atrial flutter and fibrillation (atrial fibrillation in the horse is not rare—Roos). Records illustrating some of these conditions will be shown.

Measurement of total cerebral blood flow in the monkey (Macacus rhesus).

P. R. Dumke (by invitation) and C. F. Schmidt. *Laboratory of Pharmacology, University of Pennsylvania, Philadelphia.*

The animals are anesthetized with nembutal, the basilar and external carotid arteries are ligated, heparin is injected intravenously at frequent intervals, and the volume of blood flowing through both internal carotids is measured by timing the interval required for an injected air bubble (later removed by a suitable trap) to traverse a measured distance in a glass tube interposed between the cardiac and cephalic ends of the carotids. Control observations indicate the method to be accurate over the range of flows here encountered. Terminal injection experiments show that the measured blood did not escape to any extracranial tissue and that only insignificant amounts of blood could have reached the brain by other channels. In 14 experiments so far performed the volume of blood flow at the start (under "normal" conditions) ranged from 0.36 to 0.77 cc. per gram of brain (above the basilar ligature) per minute and the highest flow encountered was 1.11 cc./ gram/ min. (during severe anoxemia). Spontaneous decreases in flow, progressing even to the point at which symptoms of acute cerebral anemia were elicited, were occasionally seen; these could be remedied (usually permanently) by aminophyllin. Cervical sympathetic stimulation had no significant effect on flow. Adrenalin injected into the carotid blood regularly caused marked reduction in cerebral blood flow. Ergotamine and benzedrine had similar though less intense effects. Caffeine, histamine, and nitrites increased the flow, but aminophyllin was the most effective of this group. Oxygen inhalation usually caused a slight and probably insignificant decrease in the flow; anoxemia caused a marked increase in experiments in which it was well tolerated. CO_2 inhalation usually caused some increase in flow but the effect was not nearly as great as might be expected from observations made on other species. Metrazol caused a pure increase in flow even when given in amounts sufficient to cause violent convulsions. Further experiments are being made.

The utilization of the lower fatty acids by normal and eviscerated animals.

J. A. Dye and Roger W. Marsters (by invitation). *Department of Physiology, Cornell University, Ithaca, N. Y.*

The sodium salts of acetic, butyric, caproic, caprylic and capric acids were injected intravenously into normal and abdominally eviscerated dogs.

All animals were fasted, nembutalized and bilaterally nephrectomized. Blood samples were taken immediately before and at half-hour intervals after the injections. Blood fatty acid determinations were made using a modified Friedemann method (J. Biol. Chem. **123:** 161, 1938), total acetone bodies were determined by Ravin's method (J. Biol. Chem. **115:** 511, 1936). Before titrating the blood filtrate distillates for fatty acids, the solutions were aerated with carbon dioxide-free air for five minutes to free them of carbonic acid. Recovery values from blood were 94 per cent for acetic acid and 98–100 per cent for each of the others.

Control blood samples gave from 0 to 7 mgm. per cent of volatile fatty acids. Non-injected control animals maintained constant volatile fatty acid titration values and extremely low fasting blood acetone body concentrations. With the liver intact, each of the fatty acids studied is utilized rapidly and completely. In each case also acetone bodies were produced though in small quantities in the case of acetic acid. The utilization values for eviscerated preparations were approximately 60, 42, 42, 47 and 20 per cent for acetic, butyric, caproic, caprylic, and capric acids respectively as compared with those for normal animals. In the absence of the liver, however, there was no demonstrable production of acetone bodies in any case. Preliminary experiments with acetic acid show that the metabolism, as measured by the oxygen consumption, is increased 100 per cent or more by injections of this acid. The energy metabolism of fatty acids is being studied further.

The effect of ergotamine on the glucose tolerance curve. G. S. Eadie, A. M. Hughes (by invitation) and Dorothy Webster-Martin (by invitation). *Department of Physiology and Pharmacology, Duke University School of Medicine, Durham, N. C.* (Read by title.)

The inhibitory action of ergotamine on alimentary hyperglycemia has been explained in various ways. Pollak (Naunyn-Schmiedebergs Arch. **140:** 1, 1929) based his theory partly on the finding that the extent of absorption of glucose from tied-off intestinal loops was not affected by ergotamine in anesthetized rabbits, and postulated an action of the drug on the liver. On repeating these experiments using pentobarbital anesthesia we found that while we could confirm the absence of an effect on absorption, we could not obtain the typical effect on the blood sugar owing to the antagonistic effect of barbiturates on ergotamine. Turning to unanesthetized animals we introduced the sugar by stomach tube and were able to show that the height of the blood sugar curve was closely related to the amount of glucose disappearing from the stomach as determined by killing the animal and estimating the glucose content of the stomach (the rest of the tract contained only traces of glucose). The effect of ergotamine therefore is to delay the emptying of the stomach. Since the effect on the blood sugar is also present when the glucose solution is placed directly in the duodenum (the end of the tube was shown by X-rays to be there) it is probable that the motility of the duodenum and upper part of the intestine is also of importance.

Relation of the conditioned and conditioning mechanisms. William Eccher (by invitation) and E. A. Culler. *University of Rochester, Rochester, N. Y.* (Read by title.)

It is well known that a conditioned response (CR), when not reinforced, tends to extinguish; but the present report shows that the same CR which tends to extinguish when kept in the secondary (conditioned) role, tends to persist unimpaired when given the primary (conditioning) role. A number of cats were trained to step forward with the right hindleg in reply to a bell, failure to respond being punished by shock-to-paw. After 100 per cent conditioning had been reached (25 CR in a work-period of 25 trials), the animals were divided into three groups. In group I the bell was presented alone, the paw-shock being discontinued. In group II the training-procedure was continued; that is, the animal was shocked upon failure to react to bell. With group III the shock was discontinued, and the CR to bell was now used for conditioning the animal to a previously neutral stimulus (800-cycle tone).

Results may be summarized as follows. 1. As would be expected from earlier work, omission of the reinforcing shock leads to gradual, and eventually total, extinction of the CR to bell alone. 2. In group II, the CR to bell is maintained by use of paw-shock, as needed, but does not reach 100 per cent; it commonly ranges between 93 to 95 per cent. 3. In group III, where the CR to bell is used for conditioning the animal to react to the tone, the bell-response not only fails to extinguish as in group I but even maintains a higher mean level (about 98 per cent) than in group II. 4. As is usual in first-order conditioning, the CR to bell is rather deliberate, occurring just prior to the oncoming shock; but when used to reinforce the tonal response, the bell-CR becomes more brisk and vigorous. The cat seems to "fear" the bell more than the shock.

From the above it would appear that the conditioning stimulus-response is itself reinforced by the presence of the stimulus-response which is being conditioned. Each reinforces the other.

Measurements of mean blood flow by a rotameter.[1] R. W. Eckstein (by invitation), D. E. Gregg, A. Rotta (by invitation) and J. T. Wearn.
Department of Medicine, Western Reserve University, Cleveland, O.

This instrument, used commercially to meter fluids and gases, has been adapted to measure mean blood flow in various body regions of the anesthetized dog. Possible effects on the accuracy of the rotameter induced by variations in stroke volume, heart rate, pattern of the flow curve (determined by the orifice plate meter, Gregg and Green, Am. J. Physiol. **130:** 114, 1940) together with alterations in viscosity and temperature of the blood have been tested in anesthetized dogs and a pump system.

Calibration in a schema under rigidly controlled conditions shows an error of about 5 per cent.

When the rotameter reading is kept constant and large changes in heart rate, stroke volume, and pattern of the flow curve are induced, the flow delivery is unchanged even when the flow curve is altered from a carotid or right coronary pattern with a high systolic and gradually declining diastolic flow, to a femoral artery pattern with a high systolic and low or zero diastolic or to a left coronary arterial pattern with a systolic back flow and high diastolic flow.

Marked differences in viscosity, temperature and specific gravity occur in different experiments and grossly affect the sensitivity of the rotameter.

[1] Aided by a grant from the Commonwealth Fund.

However, in any one experiment these variables can be rigidly controlled and either show minimal or no shifts and hence readings of the rotameter can be accurately transposed into flow values.

In actual use the rotameter can be calibrated during or at the close of an experiment 1, by comparing a series of float readings with blood flow directly measured in a graduate; 2, at a known temperature and viscosity flow can be read directly from a nomogram constructed from calibration curves obtained in different experiments in which wide ranges of these effective variables were induced by different types of anesthesia, anticoagulants and operative procedure.

The state of sensory and motor centers in patients with circulatory insufficiency and in patients with hypothyroidism. NORBERT ENZER, ERNST SIMONSON and SAMUEL S. BLANKSTEIN (introduced by A. H. Steinhaus). *Mount Sinai Hospital, Milwaukee, Wis.*

The fusion frequency of flicker was measured under standard conditions of intensity of illumination, size of area illuminated etc., in 47 normal subjects, 22 patients with circulatory insufficiency from valvular lesions and hypertension, and 13 patients with hypothyroidism. The values of both groups of patients were considerably less than the normal values; there was practically no overlapping of normal and pathological values. The decrease of the values in cardiac patients reflected the degree of decompensation. In the patients with hypothyroidism the fusion frequency was not indicative of the decrease of the metabolic rate. The maximum frequency of motor impulses measured by means of the maximum frequency of finger movements was also reduced in patients with circulatory insufficiency, in general the decrease was parallel to the gravity of decompensation symptoms. The fatigability of motor centers was increased as demonstrated by the greater drop of the motor frequency during the performance. The shape of frequency fatigue curves is often altered in such patients. Similar results have been obtained in the patients with hypothyroidism. The results explain the disposition of patients with circulatory insufficiency and with hypothyroidism to increased fatigability. In cardiac patients the principle reason for the deterioration of the state of sensory and motor centers is assumed to be the lack of oxygen in the central nervous system, in the patients with hypothyroidism the decrease of the metabolism of nerve tissue.

Modification of R in chest leads following acute and chronic muscle bundle lesions. IRVING L. ERSHLER (by invitation), SIDNEY STRINGER (by invitation) and JANE SANDS ROBB. *Department of Pharmacology, College of Medicine, Syracuse University, Syracuse, N. Y.*

Twelve dogs have been anesthetized with sodium pentobarbital, operated aseptically, a coronary branch supplying one ventricular muscle band ligated, and the animal allowed to recover. Electrocardiograms were taken before and at hourly, daily, weekly or monthly intervals after operation. The surviving animals were sacrificed at 8 months and the scars dissected for muscle bundle localization. In the three standard leads the changes previously reported by one of us as characteristic of isolated muscle bundle lesions were again obtained for each of four ventricular muscles respectively. Leads 4R, 4F and lead 2 were also recorded simultaneously.

Where the infarct involved the deep bulbospiral, the anterior left head of the deep sinospiral or the superficial bulbospiral, R of 4F was lost promptly and did not return within the eight months. On the contrary, anterolateral lesions of the right ventricle and those in the anterior papillary muscle of the left ventricle, showed only a small reduction in the R wave in lead 4F in dogs. In no instance did the R of lead 4R disappear. S-T displacements in all leads and the development of negative T waves were common.

It was again observed that infarcts of the deep bulbospiral muscle were associated with 100 per cent mortality; no animal survived over 6 hours even though kept on artificial respiration with 95 per cent O_2 and 5 per cent CO_2. Animals with deep sinospiral lesions were very ill but did survive, one was among those sacrificed, another died of pulmonary embolism 6 weeks after operation following exercise. Animals with lesions in the superficial muscles were crippled very little, if at all, after the immediate effect of anesthesia passed.

In conclusion, muscle bundle localization is again observed and it is noted that the R of lead 4F remains positive in the presence of proven infarcts involving the anterior papillary muscle and the lateral wall of the right ventricle (deep sinospiral) but disappears after certain left sided lesions.

The effect of atropine on the coronary blood flow of trained dogs with denervated and partially denervated hearts. HIRAM E. ESSEX, J. F. HERRICK, F. C. MANN and E. J. BALDES. *Divisions of Experimental Medicine and of Biophysics, The Mayo Foundation, Rochester, Minn.*

In a previous study (Am. Heart J. **19:** 554, 1940) it was shown that injections of atropine caused marked, prolonged increases in coronary blood flow and heart rate in trained dogs with innervated hearts. In the present study the response of the coronary flow and heart rate to atropine (0.1 mgm. per kilo) has been observed in dogs after the following operative procedures: 1, removal of both sympathetic chains of ganglia from the 8th or 9th intercostal space anteriorly including the stellate ganglion; 2, double vagotomy in the neck, and 3, procedures 1 and 2. The operations were done under general anesthesia employing sterile technic. The thermostromuhr units were applied to the circumflex branch of the left coronary artery at varying intervals of time following the operative procedures indicated under 1, 2 and 3. Twenty-four to forty-eight hours after application of the unit observations were begun. In the absence of the sympathetic chains as in no. 1, atropine caused an increase of 25 to 60 per cent in coronary flow and an increase in pulse rate of a similar magnitude. Atropine was without effect on the coronary blood flow and heart rate of vagotomized animals or animals with denervated hearts. Since the blood pressure is not significantly altered by injections of atropine the increased coronary blood flow is not owing to an augmented blood pressure.

Biological assays—a teaching film. HAROLD N. ETS, JOHN VAICHULIS (by invitation) and JOHN MAURER (by invitation). *Loyola University School of Medicine, Chicago, Ill.* (Motion picture demonstration.)

The film shows the assay of ergot and of digitalis by the methods given in the U. S. P. XI.

The mechanism of shock in intestinal strangulation. EVERETT IDRIS EVANS.[1] *Surgical Research Laboratories of the Harvard Medical School at the Massachusetts General Hospital, Boston.*

Shock has been produced by strangulation of a short loop of terminal ileum in the dog under Evipal anesthesia. Plasma volumes have been determined by the Gregersen dye method which has been found adequate and reliable for these shock studies. After 12–14 hours there has occurred a decrease in plasma volume of approximately 35 per cent and these animals are in varying degrees of shock, with hemoconcentration, rapid pulse and low blood pressure.

When the fluid free in the peritoneal cavity is examined, it is found to resemble closely blood plasma. When measured, it is found in amounts large enough to account for the decrease in circulating plasma. There was no evidence found to suggest loss of plasma at sites remote from the point of injury. The free peritoneal fluid of these shock animals does not appear to be toxic when given *intravenously* to a normal dog.

Hence, it is suggested that in intestinal strangulation, as in shock produced by trauma (Blalock, Phemister), plasma loss is local, and is the initiating factor in the fall in blood volume and blood pressure in this type of shock.

Effects of changes in metabolic conditions on the radiosensitivity of new-born rats. TITUS EVANS, JAMES GOODRICH and JOSEPH SLAUGHTER (introduced by J. H. Bodine). *Departments of Zoology and Radiology, State University of Iowa, Iowa City.*

New-born rats were irradiated with dosages of 300 to 3000 roentgens and the effects noted on the skin (histologically) two weeks later. It was found that animals irradiated at temperatures of 0–10 degrees C. were much more resistant to the radiation than those at room temperature. It was also found that at 30 and 35 degrees C. the injury produced was greater than that at 25 degrees C.

The effect of the temperature changes appear to be due (at least in part) to alterations produced in metabolic conditions. This conclusion is based on experiments in which the resistance was increased by preventing breathing during the irradiation. It was also found that legs and tails were more resistant if a ligature was applied during the roentgen treatment.

Desoxycorticosterone: A, its loss in the digestive tract, B, effect on water intoxication. W. J. EVERSOLE (by invitation) and ROBERT GAUNT. *Department of Biology, Washington Square College of Arts and Science, New York University, New York City.* (Read by title.)

A. We confirmed the fact (Kiuzenga *et al.*, and others) that desoxycorticosterone acetate in oil (DCA), given in a single daily dose by gavage, is ineffective in maintaining life in young adrenalectomized rats in doses (0.5 mgm./day) more than 10X those needed subcutaneously. Others have shown that such a wide discrepancy between oral and parenteral dosage does not exist for cortin and other crystalline cortical steroids. This suggested that DCA, like the chemically allied sex hormones, might

[1] Rockefeller Foundation Fellow in Surgery. Assistant in Surgery, Medical College of Virginia, Richmond.

be inactivated in the liver. This was tested by implanting pellets of DCA (mixed with cholesterol) in the spleens of young adrenalectomized rats—in which site gonad hormones are inactivated due to their immediate hepatic portal drainage. Controls received similar pellets in the kidney, subcutaneously and intraperitoneally. The pellets were of such size as barely to afford life maintenance in most but not all animals, so there was no excess of hormone involved. There was no significant difference in growth and survival between animals with pellets implanted in the spleen and their various controls. This indicates that the tissues of the liver and spleen do not inactivate DCA; and that, when given orally, it must be destroyed in the gut, since it is not detectable in the feces (Kiuzenga *et al.*).

B. Adrenalectomized animals are sensitive to the intoxicating effects of excess water, and can be protected by cortin. Swingle *et al.* have recently shown that they can also be protected by DCA. We confirmed this observation in rats, but found in addition that compared to cortin, DCA is less than one-fifth as effective in protecting against water intoxication, as it is in the maintenance of life and growth. A concentrated cortin-in-oil was used (1 cc. = 200 grams fresh gland). In life and growth maintenance 1 mgm. DCA was slightly more effective than 1 cc. cortin. (Actual m. e. d's. for 50 grams adrenalectomized rats: 0.05 cc. cortin and slightly less than 0.05 mgm. DCA.) In protecting against excess water, 5 mgm. DCA was closely equivalent or slightly inferior to 1 cc. cortin. (Actual m. e. d.'s approximated 0.2 cc. cortin and 1 mgm. DCA.)

The relation of injury potentials in heart muscle to other electrical and to mechanical events.[1] J. A. E. Eyster and Harold Goldberg (by invitation). *Department of Physiology, University of Wisconsin, Madison.*

The injury activity potential (iap), recorded by means of the suction electrode applied to the ventricular surface, starts before the onset of the initial slow rise of intraventricular pressure in both the dog and turtle. The maximum of the iap occurs during the period of initial slow rise of pressure or shortly after this period ends.

A suction electrode, modified to serve also as a differential electrode has been used to compare the differential potential time curve from a region, before injury, was produced and an injury potential time curve from the same region immediately after injury was produced by suction. Records taken with this electrode show that the main peak of the differential potential time curve taken before injury coincides in time with the maximum gradient of the iap after injury. This instant is at or near the time when the iap is changing from a negative to a positive potential value with respect to the potential of uninjured inactive muscle.

In the ventricle of the turtle, the iap develops fully within one cardiac cycle following the application of suction to a local surface region. If the suction is applied during diastole, the first change is in the negative direction and the injury resting potential (irp) develops rapidly to its full value. If the suction is applied during ventricular systole, the first change is in the positive direction and the iap develops rapidly. The magnitude of the iap in this cycle is greatest if the suction is applied early in systole and decreases in magnitude as the onset of suction occurs progressively later in the systolic period.

[1] Supported in part by a grant from the Wisconsin Alumni Research Foundation.

The influence of variations in environmental temperature and of fever on the vago-insulin and sympathetico-adrenal systems. J. FELDMAN (by invitation) and E. GELLHORN. *Department of Physiology, College of Medicine, University of Illinois, Chicago.*[1]

Normal (A), adreno-demedullated (B), and adreno-demedullated-vagotomized (C) rats were exposed to cold and heat and the effect of these procedures on the vago-insulin (V.I.) and sympathetico-adrenal (S.A.) systems was studied. It was found that cold causes hyperglycemia in A, hypoglycemia in B, and no change in blood sugar in C. Heat produces in A and B, a hypoglycemia which is however greatly delayed in A; during this time the blood sugar rises slightly. No significant change in blood sugar occurs in C. In group (D) [vagotomized rats with normal adrenals], heat causes hyperglycemia. The experiments show that heat and cold cause an excitation of both V.I. and S.A. systems, the former reacting predominantly to high, the latter largely to low environmental temperature.

Typhoid para-typhoid vaccine (Abbott 0.07 cc./kgm. subcut.) produces hyperglycemia in A, hypoglycemia in B, and no significant change in blood sugar in C. Group D reacts with a more prolonged hyperglycemia than A due to the elimination of the vago-insulin effects. Apparently foreign protein act on the centers of the V.I. and S.A. systems with a predominance on the latter in the normal animal. It is noteworthy that the study of the body temperature gives further evidence of the excitement of the para-sympathetic and sympathetic systems. The temperature rises in group A and D, and in the latter the rise is prolonged. A fall in temperature occurs in group B, no change in group C. It is assumed that the changes in temperature are due to the liberation of adrenalin in A and D and to insulin in B.

The action of propylcephaeline disulfamate on the blood cells. NELLO M. FELICELLI (by invitation) and HAROLD N. ETS. *Loyola University School of Medicine, Chicago, Ill.* (Read by title.)

Propylcephaeline disulfamate was injected intramuscularly into white mice in doses of 0.009 mgm. per gram of mouse on three alternate days. Blood was obtained from the tail at the time of making the injections and standard procedures were used in making blood counts.

There was a tendency for a decrease in the number of red blood cells following injections, but the total number of white cells showed a very marked decrease. The differential count revealed that the polymorphonuclear leucocytes suffered the greatest loss, but there was a tendency for the immature neutrophiles to increase.

The lymphoid cells showed an increase and there was an increase in the number of immature lymphoid cells.

The physical symptoms noted were: a decrease in body weight after each injection; edema of the conjunctiva associated with a discharge; and a cyanosis, as evidenced by a faster respiration, color of the mucous membranes, and the color of the blood after it was drawn.

We are indebted to Dr. Bertha Van Hoosen for the propylcephaeline disulfamate used in this work, and to Dr. Paul G. Schmitt for checking some of the differential counts.

[1] Aided by a grant from the John and Mary R. Markle Foundation and W.P.A. Project 30278.

Heparin and natural antiprothrombins in relation to prothrombin assay. JOHN H. FERGUSON and ANTHONY J. GLAZKO (by invitation). *Department of Pharmacology, University of Michigan, Ann Arbor.*

Clotting-times can be true measures of thrombin (and prothrombin) only when the standardized test conditions include complete control of all inhibitory mechanisms. An assay technique is presented to show that prothrombin preparations give an excess yield of thrombin on dilution. Progressive and immediate antithrombins are ruled out and the excess of thromboplastin ensures that it is not a variable. The explanation is in the effect of dilution on the action of a naturally-occurring *antiprothrombin.* Heparin is an antiprothrombin in the first phase of coagulation and this action consists of 1, slowing the rate of thrombin formation (probably due to inhibition of thromboplastic enzyme), and 2, lessening of amount (effectiveness) of thrombin formed (due to physico-chemical changes of prothrombin [protein] itself). Although not requiring a co-factor in the systems used, these heparin effects are greatly enhanced by an "albumin" preparation in amounts which show little proantithrombic (second phase) action. Notwithstanding the difficulties of diluting out the antiprothrombic action of the natural factor and of added heparin, it is shown to be possible quantitatively to "recover" prothrombin mixed with very dilute, defibrinated, blood plasma.

Experiments on the water balance of the dolphin. E. S. FETCHER, JR. (introduced by M. B. Visscher). *Department of Physiology, University of Minnesota, Minneapolis.*

Experiments to discover possible special mechanisms of water regulation were carried out on two unanesthetized *Tursiops truncatus.* The dolphins were placed in a trough under running showers. Control samples of blood, urine, feces, and saliva were taken. Two liters of 0.55 M sodium chloride were given by mouth; this amount and concentration is comparable to a meal of marine invertebrates, including metabolic water. The excreta were collected for several hours, analyzed for chloride and the volumes measured. The two experiments checked well. The following conclusions are possible:

1. Serum chloride was very constant: the maximum difference was 9 per cent before and after ingestion of the salt solution.

2. The kidney can secrete a high concentration of chloride, 0.57 M in one experiment, but probably for short periods only, and not normally (Fetcher, Quart. Rev. Biol. **14**: 451, 1939).

3. The saliva played no significant rôle in water regulation.

4. Feces had essentially the same chloride concentration as the blood. Consequently if a hypertonic solution is ingested, sodium chloride is selectively absorbed by the intestine (as with other mammals) a process detrimental to the water regulation.

5. Largely because of water loss from the intestine total salt excretion was relatively low. Thus, at 500 minutes 81 per cent of the water had been excreted and only 53 per cent of the salt, leaving in effect a 1.52 M solution of the salt to be excreted.

6. The experimental animals had not reached a normal state at the end of the experiments, unless water was absorbed through the skin, and it and salt were not excreted; such absorption is unlikely because of the struc-

ture of the skin. Selective absorption of water in the buccal cavity would provide a source of water unavailable to the animals in these experiments.

Action of potassium and strophanthin on the stomach of the turtle. DOROTHY FETTER (introduced by F. H. Pike). *Department of Industrial Hygiene, Delamar Institute of Public Health, Columbia University, New York City.* (Read by title.)

The slider terrapin was used. The spinal cord was cut and the plastron removed. A small rubber tube was tied in the oesophagus.

Generally, no movements were observable in the stomach, but strong constriction of the pylorus or of circular muscles lower down in the intestine were frequently present. A nematode, Camallanus trispinosus Leidy 1851, and an acanthocephalid, Neoechinorhynchus Leidy 1851, were present in such animals. (Identification by Dr. James Culbertson and Prof. L. J. Thomas.) The liver was usually yellowish instead of red in infested animals.

Movements of the stomach followed introduction of 20 cc. of water into the stomach of an infested animal, but might not appear in others. Contractions always followed excitation of the vagus, the left being more effective on the stomach, and the right on the heart.

Introducing 2 cc. M/10 KCl into the stomach always evoked movements in infested animals, and usually in others. Slightly greater amounts always induced contractions. One to two cat units of strophanthin in the stomach reduced movements of the stomach and constriction of pylorus and intestine, either before or after potassium.

Following introduction of potassium into the stomach, excitability of the vagus, judged by effects on both heart and stomach, increased. A heart insensitive to left vagus under control conditions might stop. And a stomach, insensitive to the right vagus, would contract. About two hours after administration of strophanthin, excitability of the vagus would fall, and a heart insensitive to left vagus under control conditions would again become insensitive to it. Currents of greater intensity than were necessary at the height of the potassium effect became necessary for both heart and stomach as the strophanthin effect appeared. The potassium effect was always less marked, and the strophanthin effect more marked, in animals relatively free from parasites and with a red liver, than in infested animals.

Peripheral vascular responses to the ingestion of food. SIDNEY M. FIERST (by invitation) and DAVID I. ABRAMSON. *May Institute for Medical Research, The Jewish Hospital, Cincinnati, O.*

Since it has been shown that the cardiac output is significantly increased during digestion, the question arises as to whether or not the peripheral blood flow is similarly affected. Therefore the influence of a meal of carbohydrate or protein upon the rate of blood flow through the hand, forearm and leg was studied by means of the venous occlusion plethysmographic method.

Control blood flow readings were obtained upon seven normal subjects in the post-absorptive state, and then approximately 400 calories of protein or carbohydrate were ingested; the experiment being continued for the subsequent two to four hours. The reaction to both types of food was frequently studied in the same individual.

In every instance the rate of blood flow through the hand was somewhat increased by a protein meal. The maximal effect was observed one and one-half to three hours post-prandially. Following a carbohydrate meal, however, no definite augmentation in flow through the hand took place.

In the forearm and leg, an increase in blood flow was observed after a protein meal if the experiment lasted for more than two and one-half hours. When a shorter period was utilized no change was recorded. A carbohydrate meal produced no significant effect upon blood flow in the forearm and leg.

Following ingestion of both carbohydrate and protein, a definite increase in pulse rate was frequently noted. Changes in blood pressure were also present, the greatest effect being produced by protein intake. In more than half the cases an increase in pulse pressure, as a result of either an elevated systolic or a decreased diastolic pressure, was observed.

An increase in the rate of oxygen consumption followed the ingestion of both types of food, but there was no obvious correlation between these changes and the alteration in peripheral blood flow. This was particularly true in the case of the carbohydrate meal, following which the metabolic rate in some instances was increased as much as 21 per cent without any significant augmentation in peripheral blood flow.

Effect of thiocyanate on intestinal secretion. K. FINK (by invitation) and E. S. NASSET. *Department of Vital Economics, University of Rochester, Rochester, N. Y.*

A study was made of the effect of thiocyanate on ileal and colonic secretion in the dog. After the administration of sodium thiocyanate by stomach tube, the volume, total enzyme (amifase, invertase and peptidase), and mucoprotein were increased as much as two to four times the control values. The concentration of chloride and carbon dioxide in the juice was not significantly altered.

Thiocyanate had no demonstrable effect upon the oxygen consumption of intestinal mucosa.

Progress in the purification of enterocrinin. R. M. FINK (by invitation) and E. S. NASSET. *Department of Vital Economics, University of Rochester, Rochester, N. Y.*

Using an improved method of bioassay, a quantitative study of the increase in potency and the yield of enterocrinin obtainable by a large number of fractionation procedures has been carried out. A combination of some of the better procedures has resulted in a 300-fold increase in potency and removal of all but traces of the secretin and vasodilatory substances present in the crude extracts. The most potent preparation so far obtained significantly increased the rate of secretion of succus entericus when a dose of 80 micrograms was administered intravenously to a 20 kgm. dog.

The renal clearances of hippuric acid and pyridone derivatives.[1] NORMA FINKELSTEIN (by invitation), LUCY M. ALIMINOSA (by invitation) and HOMER W. SMITH. *Department of Physiology, New York University College of Medicine, New York City.*

[1] Aided by a grant from the Commonwealth Fund.

Using appropriate colorimetric methods of analysis, or alternatively a Hilger two-beam quartz spectrograph, we have compared the renal clearances of 1, o-hydroxy hippuric acid; 2, p-hydroxy hippuric acid; 3, p-amino hippuric acid; 4, p-acetylamino hippuric acid, and 5, α-pyridone-N-acetic acid with 6, diodrast; 7, hippuran or 8, iopax in the dog. (Those compounds other than the iodine derivatives were kindly synthesized for us by Dr. Kenneth C. Blanchard.) It is concluded that the clearances of (2), (3), (4), (6) and (7) are identical within the limits of experimental error. (The excretion of these substances cannot be examined by simultaneous clearances, since they mutually depress the tubular excretion of each other. The problem requires the observation of *successive* clearances under conditions where the renal blood flow and filtration rate are as constant as possible.)

The demonstration that five substances have identical (and nearly maximal) clearances does not prove that the overall renal extraction ratio is 1.0, for any renal blood not available for clearance in respect to one substance would in principle not be available for clearance in respect to the others. This identity does show, however, that the limiting factor in the extraction ratio of any one of these substances is the distribution of blood to excretory tissue, rather than some factor in diffusion or tubular activity, as is the case with phenol red, the clearance of which is substantially below the clearance of this group of compounds.

The investigation is being extended to the comparison of the maximal rate of tubular excretion and related problems, the *in vivo* penetration of red cells and the extraction ratio in the explanted kidney in the dog, and to a comparison of clearances in man.

Studies concerning the relation between drug activity and exposure to low atmospheric pressure (high altitude). ERNST FISCHER. *Department of Physiology and Pharmacology, Medical College of Virginia, Richmond.*

Mice or rats exposed daily for two hours to a low atmospheric pressure simulating an altitude of 22,000 feet, show after 10 to 14 days in comparison with control animals a retardation in weight gain, an increased R.B.C. count, and an increased resistance to the lethal effect of severe anoxia produced by exposure to altitudes above 30,000 feet. The susceptibility to morphine and to sulfanilamide of the exposed animals is distinctly diminished for several days after the last exposure. There is experimental evidence that only the change in sulfanilamide susceptibility can be accounted for by the observed difference between the exposed animals and the controls.

Immediately after a single exposure to 22,000 feet, morphine susceptibility is temporarily decreased, while sulfanilamide susceptibility is strongly increased by a single two hours exposure to 22,000 feet shortly after administration of the drug.

Gitalin (5 to 10 γ/gm.) has a prophylactic effect against the lethal effect of high altitude above 30,000 feet. The protection afforded is at its height for a period from 16 through 30 hours after injection. The protection afforded becomes more distinct with increased altitude. Apomorphine (10 γ/gm.) has an immediate but weaker protective action, also distinctly depending on the altitude. The gitalin and the apomorphine prophylaxes are additively synergetic. Simultaneous records of respiration and E.K.G. of protected as well as of unprotected rats during exposure to high altitudes

indicate that apomorphine delays the anoxemic paralysis of the respiratory center, while gitalin stabilizes the heart rhythm and delays the anoxemic damage of the heart muscle as expressed by the changes in voltage and shape of the T-waves.

A mechanism of narcosis suggested by the effects of narcotics on several types of cell. KENNETH C. FISHER. *Department of Biology, University of Toronto, Toronto, Canada.*

Attempts have been made recently in this laboratory to describe the relation between the concentration of a narcotic and its effect on the respiration of various cells by means of an expression of the law of mass action. New data obtained, as well as some found in the literature, suggest that the action of ethyl urethane is due to its effects on two respiratory systems. The sum of these two parallel systems constitutes the normal oxygen consumption of the cells. They are differentiated on the basis of their sensitivity to the narcotic and on their different reaction with it.

Inhibition of the more sensitive of the two systems may be complete, with little or no effect on the second, and with relatively much less than complete inhibition of the total oxygen consumption. If the activity of cell division or of light production is associated with the integrity of this more-sensitive respiratory system, as seems to be the case, then one can readily account for the common observation that narcotics affect function faster than total respiration as the concentration is increased.

The quantitative effects of certain narcotics other than ethyl urethane which we have examined, do not always suggest upon gross examination that the narcotic is affecting two systems. If, however, it be postulated that the affinities of the two systems are not in the same ratio for all inhibiting substances, then all effects so far observed on yeast are completely consistent with the view that narcotics in general actually inhibit two distinct respiratory systems. This postulation of course provides a mechanism for the differences between the minimum lethal and the minimum hypnotic effects of different narcotic agents.

Studies on the effect of raw pancreas fed to completely depancreatized dogs. J. B. FLANAGAN (by invitation) and H. C. STRUCK. *Department of Physiology, College of Medicine, University of Illinois, Chicago.*

Completely depancreatized dogs have been maintained for periods up to a year. No constant effects of feeding large amounts of fresh, raw beef pancreas to these animals could be shown, either on biopsies of livers or on insulin requirements of the animals. There was a questionably significant decrease in fatty degeneration of livers of depancreatized dogs given large amounts of vitamin D in addition.

Respiratory enzymes and histologic structure of the developing cerebral cortex of the fetal pig. JOSEFA B. FLEXNER (by invitation), LOUIS B. FLEXNER and WILLIAM L. STRAUS, JR. (by invitation). *Department of Embryology, Carnegie Institution of Washington, Baltimore, Md., and the Department of Anatomy, The Johns Hopkins University.* (Read by title.)

Estimations have been made of oxygen consumption, cytochrome-cytochrome oxidase activity and cytochrome oxidase activity of the cerebral

cortex of the fetal pig from about the thirty-fifth day of gestation until term (114 days). Sliced or minced cortex has a Q_{O_2} of about 6 until the ninetieth day of pregnancy; then gradually rises to the value of 8.5 found at term.

The curve describing the change of Q_{O_2} with gestation age is apparently in no way related to the curve describing the change of cytochrome-cytochrome oxidase activity as measured with p-phenylenediamine. Minced or sliced cortex shows no cytochrome-cytochrome oxidase activity for approximately the first half of gestation; then rapidly rises during the period of a week to about one-fifth of the value of adult cerebral cortex; and after this initial increase, gradually doubles in intensity to term. Ground cortex shows slight cytochrome-cytochrome oxidase activity during the first half of gestation and a sharp increase following this period. The results obtained on cytochrome-cytochrome oxidase activity with minced or sliced brain consequently appears due to two factors. The first is an increase in permeability to the reagent, p-phenylenediamine, at the end of the first half of gestation. The second and quantitatively more important, is a sharp increase in cytochrome-cytochrome oxidase activity.

Cytochrome oxidase activity, estimated with p-phenylenediamine in presence of an excess of cytochrome C, was found to be constant throughout that period of fetal life which was investigated. This suggested that the low cytochrome-cytochrome oxidase activity found in the first half of the gestation period was due to low concentration of cytochrome C. This conclusion was substantiated by spectroscopic examination of suspensions of fetal cerebral cortex.

At all stages of development, the oxygen-uptake of the cortex was diminished by more than 90 per cent by 0.001 M cyanide.

At the end of the first half of gestation and at the time of the sharp increase in cytochrome-cytochrome oxidase activity, there was a general and marked increase in the size of the nerve cell bodies and in the quantity of their Nissl substance.

Electroencephalographic studies during acute experimental increases of intracranial pressure. FRANCIS M. FORSTER[1] (by invitation) and LESLIE F. NIMS. *Laboratory of Physiology, Yale University School of Medicine, New Haven, Conn.*

Four cats, four dogs and two monkeys were studied during periods of increased intracranial pressure induced by Cushing's technique. Blood pressure and electroencephalograms were continuously recorded. The factor of importance in inducing electroencephalographic changes was found to be not the height of the intracranial pressure but its relation to the blood pressure. A diminution in frequency and amplitude occurred when the intracranial pressure approached within 40 mm. Hg of the blood pressure and complete obliteration of the electroencephalogram occurred when the intracranial pressure equaled or exceeded the blood pressure. Blood pressure responses could be induced in the absence of cortical electrical activity.

Pharmacological observations on two water-soluble vitamin K-like substances. R. H. K. FOSTER, JAMES J. SMITH (by invitation) and A. C.

[1] Rockefeller Foundation Fellow, 1940–1941.

IVY. *Pharmacology Laboratory, Hoffmann-La Roche, Inc., Nutley, N. J. and the Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Hypoprothrombinemia in rats was produced by bile duct obstruction or by feeding petrolagar. Quick's prothrombin time method (PT) was employed and blood obtained by cardiopuncture under ether anesthesia. The mean PT on 144 normal rats was 22.9″ and on 60 deficient rats 51.5″. Four γ per rat of tetrasodium 2-methyl-1,4-naphthohydroquinone diphosphoric acid ester (N-123) or of 2-methyl-1,4-naphthohydroquinone-3-sodium sulfonate (MNSS) lowered the PT to 32″ within 24 hrs. Two γ lowered it to 38″ and 43″ for N-123 and MNSS respectively. On a molecular weight basis N-123 is twice as active as MNSS.

In rats the subcutaneous LD50 of N-123 was 610 mgm/kgm. and of MNSS 175 mgm/kgm. Deaths occurred in 12 to 140 hrs. Post mortem findings for both substances consisted in moderate gross or petechial hemorrhages and congestion in various organs. Slight microscopic changes occurred in the kidney. In rats receiving 10 doses of 100 mgm/kgm. of N-123 during 14 days there were no microscopic changes and the treated rats gained as much weight as controls. With 18 similar doses administered during 18 days there was a significant loss of weight. A slight loss of weight occurred with 25 and 50 mgm/kgm. doses of N-123 or MNSS given daily for 18 days. 5 mgm/kgm. of either substance resulted in a gain in body weight significantly greater than controls.

Rabbits receiving subcutaneously up to 20 doses of 100 mgm/kgm. of N-123 developed a low grade anemia with complete recovery starting even during the treatment. Four daily doses of 150 mgm/kgm. of N-123 caused a profound aplastic anemia and 50 per cent mortality.

Rats receiving repeated high doses of either substance developed persistent yellow-brown pigmentation of the fur, symmetrically distributed about the head and body in discrete areas. In rabbits receiving N-123 a similar pigmentation also developed but the color approached a pink-orange.

In pentobarbitalized dogs 20 mgm/kgm. of either substance caused no effect on BP. With 30 mgm/kgm. N-123 caused a slight rise and MNSS a slight fall or no effect. Forty milligrams per kilogram of N-123 caused a marked rise in BP with increased respiration and awakening. In cats under alurate these effects appeared with one-fourth the dose.

Gastric secretion under conditions of orthostatic handicap to the circulation.

ELIZABETH BROGDON FRANSEEN (introduced by Charlotte Haywood). *Department of Physiology, Mount Holyoke College, South Hadley, Mass.*

Fixation of humans in a standing position is known to induce orthostatic collapse. The squeezing action of anti-gravity muscles incidental to postural sway over a stationary base has been shown to avert such circulatory failure (Brogdon and Hellebrandt, Am. J. Physiol. **129:** P318, 1940). This study has been concerned with further observations of the cardiovascular response to standing with particular reference to concomitant gastric activity.

A test meal, 50 cc. of 7 per cent alcohol, was introduced into the fasting stomach through a duodenal tube 15 minutes after the onset of standing. Samples of the gastric contents were withdrawn at 15 minute intervals

for fractional analysis of free and total acidity. The subjects were healthy young women. They easily tolerated 75 minutes of standing when free physiological sway was allowed. Heart rates taken at minute intervals did not exceed 95 per minute. It was not always possible to stand through the secretory cycle when held immobilized by extrinsic supports. Under that condition the heart rate rose progressively to 110 or higher, syncope threatened, and the subject was returned to the horizontal.

The secretory response of the normal stomach is highly variable even in the tube-trained subject (Hellebrandt and Brogdon, Am. J. Dis. and Nutrition **2:** 402, 1935). However, the acidity values obtained throughout and following the periods of fixed standing which were associated with exaggerated cardiac response are clearly depressed below the range of those of recumbency. With free postural sway, the secretory curves are indistinguishable from those of recumbency. It is suggested that vasoconstriction in the splanchnic region, while temporarily maintaining an adequate cerebral circulation during rigid standing, may be so extreme as to impair visceral function. Postural sway serves as a circulatory aid so effectively that the gastric cells are forced to suffer little interference with their work through a reduction in their blood supply.

The interference in the absorption of inorganic phosphorus by aluminum hydroxide: Its use in children with chronic renal insufficiency. SMITH FREEMAN and WILLIE MAE FREEMAN (by invitation). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Studies carried out on normal dogs have demonstrated the effectiveness of aluminum hydroxide in interfering with the absorption of inorganic phosphorus. One hundred twenty cubic centimeters of 5 per cent aluminum hydroxide daily will reduce the urinary excretion of inorganic phosphorus to approximately half of its normal value in dogs receiving a diet containing liberal amounts of calcium and phosphorus (1 gram each daily). Similar results have been obtained on normal adult male human beings who were receiving a diet containing approximately 2 grams each of calcium and phosphorus daily. By the use of a low phosphorus diet in conjunction with 120 cc. of 5 per cent aluminum hydroxide daily it was possible to produce a negative phosphorus balance in a child with "marble bones."

Reducing the phosphorus absorption, by restricting its intake or by using aluminum hydroxide to interfere with its absorption, resulted in a decreased urinary excretion of inorganic phosphorus in 3 children with chronic renal insufficiency. In all three children the initally high serum inorganic phosphorus content of the serum was reduced by restricting the phosphorus absorption. An inverse relation between the serum calcium and inorganic phosphorus was observed frequently in these children. A reduction in some of the clinical manifestations of renal insufficiency was observed with the decrease in serum inorganic phosphorus.

The fluorescence of chlorophyll in plants and its relation to the induction period of photosynthesis. C. S. FRENCH, T. T. PUCK and J. FRANCK (introduced by E. M. K. Geiling). *University of Chicago, Chicago, Ill.*

Both the fluorescence outburst and the induction period of photosyn-

thesis which occur at the beginning of illumination of a leaf are due to the fact that the catalyst involved in splitting oxygen off of peroxides becomes inactivated in the dark. As a result, the photochemically produced peroxides accumulate and, by oxidation, form substances which inhibit photosynthesis and increase the fluorescence yield. These substances are removed by a thermal reaction, so that the fluorescence intensity at any moment is determined by the balance between these two reactions. As photosynthesis proceeds, the catalyst in question is reactivated until it can remove the peroxides rapidly enough to avoid the oxidation processes. Evidence is presented by experiments wherein the light intensity is suddenly decreased after a very short, intense illumination, so that the rate of disappearance of the substances causing the resulting fluorescence outburst can be followed. Their lifetime can also be estimated by measuring the initial fluorescence intensity after dark periods of varying duration. At room temperature, the half-life of these substances in the dark is 1.7 seconds. HCN, an excess of CO_2 or low temperatures decrease the rate of the thermal reaction removing these substances, and so prolong the time of the fluorescence decay, as well as of the induction period.

Studies in renal vein occlusion.[1] L. FRIEDBERG (introduced by L. N. Katz). *Cardiovascular Department, Michael Reese Hospital, and Department of Physiology, University of Chicago, Chicago, Ill.*

Experimental studies in hypertension have shown that renal ischemia plays an important etiological rôle. It appeared possible that a state of relative renal ischemia might be produced by increasing the venous pressure within the kidney and that hypertension might result from this procedure. Partial occlusion of one renal vein in the rabbit has led to a transitory hypertension. Results in dogs have been inconsistent. In the present study, trained unanesthetized dogs were used, the blood pressure being determined by the Hamilton manometer. Occlusion of the renal veins was accomplished by means of linen ligatures under nembutal or ether anesthesia.

Moderate transitory hypertension was observed after partial occlusion of one renal vein. This was more evident when the animal was contralaterally nephrectomized. Collateral venous circulation developed almost immediately, and this probably led to the dissipation of the hypertension. The hypertension in such dogs lasted three or four days, the blood pressure then returning to its control level. Post-mortem examination revealed a vast collateral supply to the kidney. Reoperation and occlusion of most of the collateral supply led to a more persistent hypertension in one dog.

Complete occlusion of one renal vein was not accompanied by hypertension. The kidney was severely damaged by this procedure, however, since removal of the normal kidney resulted in uremia.

Complete occlusion of both renal veins was followed by anuria and uremic death in a few days without the development of hypertension.

Relation of the cervical sympathetics to anterior pituitary gonadotropic activity in the rat. H. B. FRIEDGOOD and S. BEVIN (by invitation). *Harvard Medical School, Boston, Mass.* (Read by title.)

Superior cervical sympathetic ganglionectomy interferes significantly

[1] Aided by the A. D. Nast Fund for Cardiac Research.

with the incidence of pseudopregnancy after glass rod or electrical stimulation of the cervix uteri. The latter induces psuedopregnancy in about 95 per cent of normal unoperated rats, whereas ganglionectomized animals become pseudopregnant after a similar stimulus in approximately 75 per cent of the cases. The gonadotropic response of the pituitary to stimulation of the cervix is thus modified experimentally to some extent by exclusion of the cervical sympathetics. One could not conclude, however, that the sympathetics normally participate in stimulation of the pituitary in the intact animal. Further evidence on this point is contained in the following observations.

NUMBER OF RATS	EXPERIMENTAL CONDITIONS	INFLUENCE ON REGULARITY OF ESTROUS CYCLES			
		None	Pseudo-pregnancy	Prolonged diestrus	Irregular cycles
11	Ether anesthesia for 10 minutes	11 (100%)	0	0	0
6	Light sodium amytal anesthesia	6 (100%)	0	0	0
43	Blank neck operations (controls)	28 (65.1%)	10 (23.2%)	3 (6.9%)	2 (4.6%)
28	Bilateral cervical sympathectomy	15 (53.5%)	11 (39.2%)	0	2 (7.1%)
53	Bilateral superior cervical sympathetic ganglionectomy or removal only of the inferior pole of both ganglia.	8 (15.1%)	34 (64.1%)	6 (11.3%)	5 (9.4%)

These significant differences in the incidence of pseudopregnancy are independent of anesthesia. They may be attributed either to indirect effects of ganglionectomy on pituitary function (e.g., vasodilatation in pituitary) or to mechanical stimulation of the ganglia and cervical chains during their removal. Ablation of the ganglia is apparently more effective in inducing pseudopregnancy than severing the chains. In conjunction with the earlier observations these data suggest that the cervical sympathetics can take part in affecting the gonadotropic activity of the anterior pituitary of the rat. It is still possible, however, that our results can be accounted for by non-specific stimulation of the pituitary through vasomotor changes.

Mediation by the small intestine of the gastric secretory depressant effect of urine extracts. M. H. F. FRIEDMAN (introduced by T. L. Patterson). *Department of Physiology, Wayne University College of Medicine, Detroit, Mich.*

Recent experiments suggest that the gastric secretory depressant substance excreted in the urine of man and dog does not act directly on the gastric glands. Experiments were performed on vagotomized dogs under nembutal anesthesia, the animals having been fasted previously for 24 hours. Gastric juice was obtained by fistula from the whole stomach and secretion stimulated by repeated subcutaneous injections of histamine (0.1 mgm. per kilo per hour). The urine extracts, which had been found

to be non-toxic, pyrogen-free and without effect on blood pressure, were administered by vein.

In a control series of 20 dogs, urine extract in effective doses inhibited gastric secretion in 90 per cent of the animals. In a second series of 16 dogs the whole of the small intestine was removed and histamine and urine extract given as in the control series. Although as much as nine times the effective dose was employed, inhibition of secretion in the enterectomized dogs was observed in only 20 per cent of the experiments. Extensive handling of the small intestine did not influence appreciably the inhibitory effect of the extract nor did the fall in blood pressure resulting from the operative procedure appear to be a responsible factor. The possibility that the inhibitory effect of urine extract on gastric secretion is mediated by the small intestine should be entertained.

The convulsant action of acid fuchsin in rats of different age periods. ALFRED FRÖHLICH (by invitation) and I. ARTHUR MIRSKY. *The May Institute for Medical Research, The Jewish Hospital, Cincinnati, O.*

In order to obtain data which may be of aid in the elucidation of the mechanism responsible for the increased susceptibility of children to convulsions, the influence of acid fuchsin was studied in rats of different age periods. Groups of from 10 to 25 rats of from 7 days of age to over 100 days of age were studied.

The subcutaneous administration of acid fuchsin in doses of 0.5 mgm. per gram of rat produced convulsions in approximately 100 per cent of animals at every age level below that of 17 days. On the 18th, 19th and 20th days of age the incidence of convulsions fell off rapidly until by the 21st day no animals responded to this dose. Increasing the dosage of acid fuchsin produced insignificant changes in the number of animals which developed convulsions.

Adult animals did not develop convulsions even when doses of as high as 3 mgm. acid fuchsin per gram was administered. However, after treatment with an injection of theophylline, even 0.5 mgm. per gram of rat resulted in the development of convulsions in about 60 per cent of adult animals.

The significance of this data as concerns the relation of age to the susceptibility to convulsions and the rôle of the hematocephalic barrier will be discussed.

The control of small blood vessels. GEORGE P. FULTON (by invitation) and BRENTON R. LUTZ. *Department of Biology, Boston University, Boston, Mass.* (Motion picture demonstration.)

In Rana pipiens, with brain and medulla destroyed, the distribution of the contractile elements of the small blood vessels in the retrolingual membrane was explored with a micro-electrode. A water immersion lens was used and cinephotomicrographs obtained. Brief faradic stimulation of non-myelinated nerves produced a diphasic response, vasodilation followed by vasoconstriction, in a limited vascular pattern. Weak stimulation generally produced only dilatation. Strong stimulation of the same nerves frequently constricted only a portion of the area originally dilated with a weak stimulus. Some nerve fibers gave only constriction or dilatation to all strengths. Each of several nerve fibers in a field may produce a

response in the same limited vascular pattern, suggesting a smooth muscle motor-unit. Faradic stimulation of small nerves frequently produced dilatation and constriction of a capillary only in the region of its origin, the response being independent of the supplying arteriole or precapillary. Such a sphincter-like mechanism may thus regulate capillary blood flow without the aid of the supplying vessel. These regions sometimes showed spontaneous independent rhythmic activity. In methylene blue preparations the capillary origins showed a few modified smooth muscle cells with branched processes. Other pericapillary cells and the endothelium itself did not respond to nerve stimulation, or to direct electrical or mechanical stimulation. The anatomically continuous perivascular nerve plexus, which is rich on the arterioles and precapillaries, but sparse or lacking on the capillaries, is physiologically discontinuous.

Reduction of carbon dioxide under anaerobic conditions in green algae. H. GAFFRON (introduced by E. M. K. Geiling). *University of Chicago, Chicago, Ill.*

With some species of unicellular green algae it was possible to demonstrate that a prolonged anaerobiosis leads to a specific change in the mechanism of the photochemical reduction of carbon dioxide. The disappearance of carbon dioxide is no more coupled with the evolution of molecular oxygen, but with the oxidation of hydrogen donors. Thus the reaction becomes similar to, if not identical with, the photochemical behavior of the purple bacteria. The course of this photoreduction in algae can be studied best with molecular hydrogen as the specific hydrogen donor. It has been known for several years that normal aerobic photosynthesis is specifically inhibited by such poisons as hydrocyanic acid and hydroxylamine. The effect of these poisons on the photoreduction with hydrogen proves that hydroxylamine exclusively inhibits the evolution of molecular oxygen under aerobic conditions, but has no influence on the reduction of carbon dioxide as such, while cyanide inhibits this latter reaction only. Anaerobic conditions also support a rapid reduction of carbon dioxide with hydrogen in the dark, provided this reaction can be coupled to an oxidation process furnishing the necessary activation energy.

Cochlear potentials elicited from bats by supersonics. ROBERT GALAMBOS (introduced by A. C. Redfield). *The Biological Laboratories, Harvard University, Cambridge, Mass.*

Cochlear potentials have been recorded from bats in response to supersonic sounds. These potentials accurately follow the frequency of the incident sound up to the upper limit of the recording apparatus (98 kc.). The maximum voltage developed remains nearly constant for frequencies between 10 and 60 kc., those between 25 and 45 kc. usually giving slightly increased voltages; the voltage at 98 kc. is about one third that at 10 kc.

Evidence that for frequencies up to 55 kc. the intra-aural muscles (tensor tympani and stapedius) function to reduce transmission across the middle ear has been gathered by measuring cochlear potentials as a function of sound intensity. The voltage developed to a moderately intense tone drops suddenly after an increase in sound intensity, and subsequent reduction of intensity is followed by a rise in voltage; this effect disappears with increase in depth of anesthesia (ether, nembutal), or upon injection

of curare. As the animal dies, the cochlear voltage first drops, then suddenly increases; the rise in voltage (2–4 min. after "death") is taken to indicate the break-down of the intra-aural muscle reflex with consequent increase in transmission across the middle ear.

Recent experiments show bats produce supersonic cries and are unable to avoid obstacles if temporarily deafened or gagged. The present study shows the bat cochlea to be sensitive to supersonic sounds. These facts taken together support the theory that flying bats hear reflections of their supersonic cries and determine the position of obstacles by auditory localization.

The author is indebted to Professor G. W. Pierce for the use of both the magnetostriction oscillator to generate supersonic tones, and the supersonic microvoltmeter to record the cochlear potentials. The following species of bats were used: *Myotis l. lucifugus* (4 ears), *M. keenii septentrionalis* (8), *Pipistrellus subflavus* (1), *Eptesicus f. fuscus* (33).

Relation between unconditioned and conditioned reflex: inhibition of CR by UR. W. Horsley Gantt. *Pavlovian Laboratory, Phipps Clinic, Johns Hopkins University, Baltimore, Md.*

Fundamentals of CR interaction have been investigated by Pavlov, and proven often similar to the laws of segmental reflex interaction as stated by Sherrington. The question of reciprocal relationship of CR–UR has been neglected. CR secretion was supposed to summate with its following UR like two allied segmental motor reflexes (Sherrington). My studies show that CR and UR however act more like antagonistic reflexes struggling for the final common path; the unconditioned stimulus immediately and completely inhibits all related conditioned reflexes. The proof is: 1, the UR is of equal magnitude whether preceded by its CR or acting without such precedence; 2, whether acting alone or concurrently with the overlapping conditioned stimulus; 3, UR is independent of the length of conditioned stimulation overlap; 4, the curves of CR and UR are independent and characteristic but the UR curve is the same regardless of preceding or simultaneous action of the conditioned stimulus; 5, there is a refractory period after the UR during which the organism is non-responsive to specifically related conditioned stimuli; 6, when UR fails to follow its CR inhibition is absent. These effects are not due to a maximum capacity secretion because it holds with even quantitatively minute URs. Even small URs can inhibit much larger conditioned reflexes, though if the unconditioned reflex is made sufficiently weak it does not completely inhibit a powerful conditioned reflex.

This inhibition is not only true for secretion and movement but for the accompanying increase in cardiac and respiratory activity, i.e., the emotional components of the conditioned reflex. Inhibition of CR by UR generally holds for the motor reflexes to pain as well as with both secretory and muscular reflexes to food. Such inhibition of conditioned reflex activity when the unconditioned reflex sets in provides a physiological resting period—involving 1, specific movements, 2, secretion, 3, emotional activity measured by cardio-respiratory responses—during which the organism is not subject to stimulation by the same conditioned stimuli to which it has just responded.

Rhythmic variations of muscular activity in normal and neurotic dogs correlated with secretion and with conditioned reflexes. W. Horsley Gantt and Wendell Muncie (by invitation). *Phipps Psychiatric Clinic, Johns Hopkins Hospital, Baltimore, Md.* (Read by title.)

The 24 hour activity was measured in 10 dogs for a period of five years using an adaptation of Richter's method applied to rats. The following facts were noted. Male dogs exhibit cycles of activity of 3–5 days' duration culminating in peaks followed by depressions. The degree of activity as well as the duration of the cycle is characteristic of the individual dog, and the pattern is maintained for long periods, perhaps throughout the dog's life. Seven pairs of normal dogs placed in adjacent outdoor cages showed parallel fluctuations in the daily activity, i.e., a high correlation factor (r), r varying from 0.28 to 0.57, mean $r = 0.47$, P (Fisher's probability factor) = 0.005. The correlation coefficient between pairs of normal dogs when the two dogs were separated, and one of the dogs placed in an indoor cage was small ($r = 0.13$). On the contrary, the correlation coefficient of a "neurotic" dog with 5 other normal dogs taken independently was negligible whether both dogs were in adjacent or in separated cages ($r = 0.08$). Furthermore there is a positive though small correlation coefficient between the amount of reflex salivary secretion to a given amount of food and the 24 hour activity ($r = 0.28$, $P = 0.05$), and an indication of a small positive correlation with the magnitude of the conditioned reflex ($r = 0.22$, $P = 0.1$). There is thus not only 1, a characteristic pattern of activity (magnitude and cycle) for each male dog, but 2, a fluctuation of activity affecting all the normal dogs dependent upon some common factor, as yet undetermined in our analyses, but possibly meteorological, as all the dogs in the adjacent outdoor cages were exposed to the weather, 3, and the individual fluctuations in 24 hour activity are roughly proportional to (i.e., positively correlated with) the physiological salivary secretion and probably the higher nervous activity, measured by the conditioned reflex, of that particular dog.

Functional organization and interrelation of cerebral hemispheres in cat.[1] Hugh W. Garol (introduced by W. S. McCulloch). *Laboratory of Neurophysiology, Yale University, School of Medicine, New Haven, Conn.*

By local strychninization of one hemisphere of the cat (*Felis domestica*) and recording electrical activity of both, the boundaries of the sensory cortex have been mapped. It has been found that while there is restricted firing from many regions within it there is no clear indication of separate subdivisions for face, arm and leg. However, there is clear indication of areas which are dissimilar in regard to the areas to which spiking potentials are propagated and these areas correspond to those plotted by Winkler and Potter in 1914 based on Brodmann's six-layer cytoarchitectonic classification. Strychninization of certain of these areas produces suppression of electrical activity of the cortex.

The origin and termination of callosal connections by location of strychnine spikes on one hemisphere from local strychninization of the other has

[1] Aided by a grant from the Knight Funds of the Yale School of Medicine.

revealed their relatively restricted origin and their projection principally to symmetrical foci.

Finally, it has been observed that suppression of electrical activity, initiated by strychninization of any suppressor areas, affects both hemispheres.

The action of acetylcholine on the heart of Limulus polyphemus. W. E. GARREY. *Department of Physiology, Vanderbilt University School of Medicine, Nashville, Tenn.* (Read by title.)

The neuromyal junctions of the anterior segments of the heart of Limulus polyphemus were not affected by acetylcholine, mecholyl or lentin. Attempts to potentiate such nonganglionated preparations with eserine failed.

The cardiac ganglion is stimulated by these choline derivatives, lentin (doryl) being by far the most effective. Acetylcholine required concentrations averaging one part in 16000, often 1 in 5000,—rarely only 1 in 30,000 was needed. Eserine produced marked potentiation of acetylcholine stimulation of the ganglion, which often was evident in dilutions of one part in a million. Stimulation involved both rate of impulse formation (cell stimulation) and height of muscular contraction (lowered cell threshold or synaptic facilitation).

Impulse transmission in the sinus venosus of the turtle heart. W. E. GARREY and C. E. KING. *Department of Physiology, Vanderbilt University School of Medicine, Nashville, Tenn.* (Read by title.)

A resurvey of impulse initiation and transmission in the sinus venosus of the turtle heart has confirmed time-honored concepts. The methods of study included high speed (64 and 128 per sec.) and ultra high speed (300 per sec.) cinematography of the contraction waves and the determination of the time relations of amplified potential changes at different loci using either unipolar or bipolar electrodes. By these methods of visualizing and measuring the progress of contraction waves, it can be stated with certainty that the normal sinus never contracts as a unit but that the contraction wave-front is initiated in and progresses radially in all directions from a small focus of initiation (pacemaker) at a rate which varies with the temperature, and eventually involves the entrant veins. The normal pacemaker is located to the right of the midline, and is usually anterior to the mouth of the coronary vein. It has been localized by other methods.

Localization of the pacemaker in the sinus venosus of the turtle heart. W. E. GARREY and C. E. KING. *Department of Physiology, Vanderbilt University School of Medicine, Nashville, Tenn.* (Read by title.)

The normal (usual) pacemaker on the sinus venosus of the turtle heart has been demonstrated to lie within an area of a few square millimeters on the ventral surface of the sinus slightly to the right of the midline and anterior to the entrant coronary vein. Its location was determined by localized stimulation of its intrinsic inhibitory mechanism.

1. A strong acetylcholine solution made into a thick paste with kaolin was "spotted" on different parts of the sinus venosus. Inhibition of rate ensued only when the drug was placed on the pacemaker-area.

2. Gentle mechanical stimulation with a camel's hair pencil-brush or wisp of cotton induced pacemaker inhibition (no extrasystole), but was ineffective elsewhere on the sinus, although rougher manipulation might stimulate intracardiac inhibitory fibers and indirectly inhibit the pacemaker.

3. Faradic stimuli delivered from fine, closely approximated electrodes could be weakened below the threshold of cardiac muscle and also below that of intrinsic vagal fibers, but still produced localized inhibition about the electrodes. Such inhibition affected the rate only when applied to the pacemaker area.

When inhibition was confined to the pacemaker, a new pacemaker developed. (cf. Meek and Eyster, 1912, 1914, 1916.)

Desoxycorticosterone and lactation. ROBERT GAUNT. *Department of Biology, Washington Square College of Arts and Science, New York University, New York City.*

Lactation deficiencies of adrenalectomized rats can be repaired completely with relatively large doses of cortin (Gaunt and Tobin). Cortin, adrenotropin or salt, as well as pituitary lactogenic hormone, must be given to initiate lactation in the hypophysectomized guinea pig (Nelson and Gaunt; Gomez and Turner). A study of crystalline cortical steroids in these responses is of interest, especially since Brownell *et al.* believe that this action of cortin is due to a separable fraction, cortilactin.

This report concerns the effect of desoxycorticosterone acetate (DCA) on the lactation of rats adrenalectomized within 24 hours after delivery. Adrenalectomy at this time permits a better lactation in untreated mothers than we previously reported when operations were done prior to parturition. All litters were reduced to 6 each. Results were as follows:

1. In untreated mothers growth of litters was sub-normal but no deaths of young occurred before 15 days and 33 per cent were raised to weaning (6 litters).

2. When the mothers received 0.3–0.5 mgm. DCA per day all the young died between the 11th and 19th days, despite large weight gains and excellent health of the mothers (8 litters). One-tenth milligram DCA gave similar results in 3 cases, but one anomalous animal lactated normally.

This would seem to indicate that DCA not only failed to support lactation but probably actually inhibited it, perhaps because it is a mammary-growth stimulating substance (Van Heuverswyn *et al.*).

3. DCA did not, however, inhibit lactation of intact rats in doses of 0.5 mgm. per day (3 litters), showing that if there is an inhibitory effect it can be prevented (at this dose level) by the normal cortical secretions.

4. This was further illustrated by giving 0.3 mgm. DCA per day plus 2 cc. Eschatin to the adrenalectomized mothers of 4 litters. All the young were raised to weaning although growth was not normal.

5. Although these results might be interpreted as indicating that DCA, due to its lack of ability to maintain a normal carbohydrate metabolism or for some analogous reason, is qualitatively incapable of sustaining lactation, such an interpretation is not necessary until the possibility that it acts as a direct lactation inhibitor is more completely ruled out.

The influence of emotional excitement on the vago-insulin system and insulin content of the blood. E. GELLHORN, A. ALLEN (by invitation),

R. CORTELL (by invitation) and J. FELDMAN (by invitation). *Department of Physiology, College of Medicine, University of Illinois, Chicago.*[1]

I. Experiments on cats. Stimulation of the hypothalamus leading to sham rage causes a fall in blood sugar in cats with denervated adrenals and liver. After additional vagotomy sham rage causes a slight hyperglycemia (sympathin) which was masked by the stimulation of the vago-insulin system. Cats with sectioning of the spinal cord at the sixth cervical level react with a hypoglycemia when confronted by a barking dog, but this effect was abolished by subdiaphragmatic vagotomy.

II. Experiments on rats. They were performed on normal (A), adreno-demedullated (B), and adreno-demedullated-vagotomized (C) animals. Emotional excitement was induced by the noise of fire-crackers and by struggle resulting from restraint. In both groups the A rats showed hyperglycemia, the B rats hypoglycemia, and the C rats no change in blood sugar. The experiments prove that emotional excitement leads to a discharge of the vago-insulin (V.I.) and sympathetico-adrenal (S.A.) systems, the effect on the latter predominating.

III. Experiments in man. The insulin content of blood of excited psychotic patients is greatly increased when assayed on the hypophysectomized-adrenalectomized rat (cf. the abstract of the paper by Allen, Feldman and Gellhorn) in spite of the absence of a significant alteration in the blood sugar of the patient. Normal individuals when excited show a rise in blood sugar and no significant increase in the insulin concentration of the blood. The experiments suggest that emotional excitement leads to a discharge over both V.I. and S.A. systems, the effect on the former predominating in psychotics, whereas in the normal the excitation of the latter prevails.

The effect of oxygen lack and inhalation of carbon dioxide on chemically induced convulsions. E. GELLHORN and L. YESINICK (by invitation). *Department of Physiology, College of Medicine, University of Illinois, Chicago.*[1] (Read by title.)

Convulsions were produced in narcotized cats by the injection of metrazol, picrotoxin, coryamyrtin, camphor, strychnine and other drugs. The blood pressure was recorded from the carotid artery and the convulsions from the hind leg. It was found that carotid sinus (C.S.) pressor reflexes are intensified under convulsions as seen by the fact that the blood pressure rise induced by adrenalin is less in the convulsant than in the non-convulsant animal. Moreover, 6 to 8 per cent O_2 inhibits convulsions rapidly in cats with denervated C.S. and divided vagi, whereas anoxia has no or very slight inhibitory effects on the convulsions of the normal animal. On the other hand, 10 to 15 per cent CO_2 abolishes convulsions within 1 or 2 minutes in the normal animal, but has only slight or no effects on the convulsions of cats which have been deprived of the buffer nerves. The peculiar reversal of the inhibitory effects of O_2 lack and CO_2 by denervation of the C.S. and vagi is thought to be due to the modifying influence of the C.S. pressor reflexes on somatic excitability. It was shown earlier (This Journal **129:** P502, 1940) that C.S. pressor reflexes inhibit

[1] Aided by a grant from the John and Mary R. Markle Foundation and W.P.A. Project 30278.

convulsions. Since these reflexes are intensified in hypercapnia but weakened in anoxia, it is to be expected that they increase the inhibitory action of CO_2 but diminish the inhibitory effects of anoxia in the normal animal. Consequently, CO_2 exerts a greater anti-convulsant effect in the normal and anoxia in the "denervated" (buffer nerves eliminated) animal.

The effects of glucose and of insulin on the metabolism of the isolated diaphragm of the rat. CHALMERS L. GEMMILL. *Department of Physiology, Johns Hopkins University School of Medicine, Baltimore, Md.*

Previous determinations (Bull. Johns Hopkins Hosp. **66:** 232, 1940; **68:** 50, 1941) of the glycogen content of the diaphragm and of the glucose utilization from the medium have demonstrated that insulin aids in glycogen deposition. These experiments have been extended by making determinations of the total carbohydrate content of this preparation, of glucose utilization from the medium and of the respiratory quotients of the muscle sections without glucose, with glucose and with glucose and insulin.

	WITHOUT GLUCOSE	WITH GLUCOSE		WITH INSULIN (1.6 U) AND GLUCOSE	
		0.2 per cent	0.5 per cent	0.2 per cent	0.5 per cent
Total carbohydrate, mgm. per 100 mgm. (wet weight) per 3 hours	0.42* 0.33	0.79	0.98	1.24	1.60
Glucose utilization from medium, mgm. per 100 mgm. (wet weight) per 3 hours		0.56	1.23	1.16	1.99
Oxygen consumption, mm.^{3} O_2 per mgm. (wet weight) per hour	0.96	0.97	1.03	0.91	0.93
Respiratory quotients	0.74	0.86	0.86	0.91	0.88

* Beginning value.

When glucose is not present in the medium, the disappearance of total carbohydrate is too small to account for the oxygen uptake. Therefore, a non-carbohydrate substrate is oxidized under these conditions. When glucose is present, the total carbohydrate content of the tissue increases, the glucose content of the medium decreases and the respiratory quotient increases. Additional glucose in the medium augments the deposition but does not change the respiratory quotient. Insulin aids also in deposition of total carbohydrate but does not affect the respiratory quotient. The oxygen consumption of the diaphragm under these various conditions is unchanged.

Summation, after-discharge and rebound. ROBERT GESELL. *Department of Physiology, University of Michigan, Ann Arbor.* (Read by title.)

Summation and after-discharge are readily demonstrable phenomena in the nervous integration of the respiratory act. They are evokable in both the inspiratory and expiratory half-centers by stimulation of sensory cutaneous nerves in which nociceptive afferents predominate. They occur

in both half-centers on stimulation of Hering's nerve in which chemoceptive fibers exert a powerful effect. They are seen on stimulation of the vagus nerve.

After-discharge is regarded as a normal and a probably highly important factor in the smooth operation of the central nervous system rather than as an abnormal phenomenon arising from unnatural conditions. After-discharge is pictured as maintaining a continuous and adequately graded nervous drive, and the electrochemical conditions producing it may be crudely compared with the supply of steam upon which the steam engine draws. By virtue of the principle of the precedence of stimulation and the associated machinery of reciprocal inhibition the "nervous power" originating in the nerve cells and their dendrites may be switched from one half-center to the other. Despite the existence of a potential power of nervous drive at both half-centers one center holds the other in abeyance and thus allows a coordinated shift in activity from one opposing half-center to the other. It is suggested that one function of the internuncial cells is to provide continuing drives by virtue of their capacity of summation and after-discharge.

The phenomenon of rebound is common in the respiratory act as it is in spinal integrations. It is thought to be intimately related to and explainable with the aid of the dual excitatory action of important respiratory receptors, the phenomenon of after-discharge, and the machinery of reciprocating interaction of half-centers.

The expiratory component in breathing during pneumothorax. ROBERT GESELL and CARL MOYER (by invitation). *Department of Physiology, University of Michigan, Ann Arbor.*

The commonly described effect of pneumothorax is a powerful inspiratory tetanus. This stands in marked contrast with the expiratory tetanic response to artificial inflation of the lungs. Our evidence, in agreement with Head, points to the fact that these diametrically opposite responses are individual reflexes; that the inspiratory contraction is supported by the collapse of the lungs and is not a negative response to a weakening of the expiratory vagal stretch reflex. Inspiratory and expiratory tetani are interrupted in each case by a progressively increasing activity of the opposing center. An increasing expiratory component during pneumothorax is readily demonstrated in the dog by recording the circumference changes of the torso. While pneumothorax always elicited a powerful inspiratory contraction, inspiratory tetanus was relatively uncommon. Instead, the frequency of respiratory movements was greater than normal. That was interpreted to mean the existence of a powerful expiratory component capable of interrupting and of balancing the newly created inspiratory drive. This expiratory component is seen to increase by the increasingly deeper expiratory excursions as pneumothorax continues. These expiratory contractions may eventually over-constrict the torso. As the expiratory component grows in power, the so-called "expiratory pauses" increase in length and breathing becomes slower than normal. This frequency effect is attributed to a disturbance in the balance of the inspiratory and expiratory drives in which the reciprocal inhibition of the inspiratory half center by the now predominant expiratory drive grows in effect. If at this stage the pleural air be removed, this super expiratory

activity is seen to continue temporarily as an after phenomenon. Despite a highly asphyxial condition of the animal (for no air was moved during pneumothorax) a prolonged apnea frequently occurs. As the after expiratory activity diminishes the frequency of breathing increases.

Three sources of expiratory drive during pneumothorax are suggested. 1. Each collapse receptor, like the stretch receptors, may be connected with both half-centers and thereby exert a dual stimulation. 2. Asphyxia adds both expiratory and inspiratory chemical drives. 3. Excessive stretching of the expiratory muscles by the augmented inspiratory contractions sets up expiratory reflex stimulation.

Proprioceptive reflex effects arising from deformations in the lungs and torso. ROBERT GESELL and CARL MOYER (by invitation). *University of Michigan, Ann Arbor.* (Read by title.)

Mechanically controlled barometric pressures producing selective inflation or deflation of the lungs or selective expansion or compression of the torso revealed deformation reflex effects on breathing.

Sustained positive pressure applied to the torso under conditions avoiding pulmonary deformation increased the frequency of breathing. The effects were slightly greater when pulmonary deformations were included.

Sustained negative pressure under similar conditions decreased the frequency of breathing. These effects were markedly greater when pulmonary deformations were included.

Rhythmic fluctuations of pressure confined to the outer surface of the torso alone readily established corresponding respiratory rhythms. The compression deformation threshold of the torso for the stimulation of inspiration as measured by the deformation pressures used was relatively uniform. Supra threshold deformation, however, prolonged and increased the intensity of inspiration.

Comparable fluctuations of pressures designed to control the deformation of the lungs alone (but unavoidably acting on both sides of the chest wall) yielded comparable results.

The prolonged expiratory response produced by pulmonary inflation classifies the stretch reflex as predominantly expiratory.

The prolonged inspiratory response frequently produced by pulmonary deflation classifies the collapse reflex as predominantly inspiratory.

The sudden interruption of an established apneustic inspiratory contraction by lung inflation is explained by the reciprocal inhibition of the inspiratory half center by the direct activation of the expiratory half center.

Demonstrable inspiratory excitatory action of the stretch reflex supports the dual excitatory action of the pulmonary stretch reflex.

Preliminary experiments point to the possibility of a similar dual excitatory action of the pulmonary collapse reflex.

The activity of single motor units in voluntary movements. A. S. GILSON and W. B. MILLS (by invitation). *Department of Physiology, Washington University School of Medicine, Saint Louis, Mo.*

Electrograms from single motor units have been recorded from normal human subjects during slight volitional efforts. If the movement be sufficiently slight and sufficiently brief, there is a single discharge of the

recording unit with each volitional effort. Between efforts the muscle may show no sign of motor activity. Similar records from a pair of opposing muscles, as for example a finger flexor and extensor, may show single motor unit discharges with each flexor or extensor effort, respectively. If a quick and somewhat more intense effort be made, there appear discharges of other units. The units of lower threshold then show an apparent reduction of latency so that there is a corresponding tendency to summation of spike potentials. If the quick movement be prolonged as a smoothly maintained tension effort, the initial responses are followed by a continued rhythmic motor-unit response, the discharge of a given unit being maintained at a rather constant rate until the tension is released. If more than one unit is responding each continues with its own rate of response so that the discharges quickly become quite asynchronous. If two or more units are brought into sustained activity successively the one of lower threshold has, in our experience, invariably continued in activity without alternation or interruption as long as the movement pattern has remained unchanged.

Quantitative effects of electrolytes and heparin on the second phase of blood coagulation.[1] ANTHONY J. GLAZKO (by invitation) and JOHN H. FERGUSON. *Department of Pharmacology, University of Michigan, Ann Arbor.*

The action of *immediate* antithrombins is sharply differentiated from that produced by *progressive* antithrombins. The kinetics of the thrombin-fibrinogen interaction are studied in the presence of various immediate antithrombins. It has already been shown by Glazko and Greenberg (Am. J. Physiol. **128:** 399, 1940) that polyvalent anions and heparin in the presence of its co-factor, both produce similar inhibitory effects on blood coagulation. Using heparin and various electrolytes, it is now demonstrated that, for a given thrombin preparation, the clotting-time is directly proportional to the concentration of immediate antithrombin. The prolongation of the clotting-time with increasing concentrations of inhibitor varies inversely with the thrombin concentration. The antithrombic effects of heparin, salts, and naturally-occuring inhibitors are compared by referring the clotting-times to the concentrations of potassium ferrocyanide required to produce the same degree of inhibition. A new principle for the assay of thrombin (activated prothrombin) is outlined, based on the increment in clotting-time produced by known concentrations of ferrocyanide. This method is compared with other methods now in use. An accelerating effect which is produced by traces of immediate antithrombins under certain conditions is also described. These results support the hypothesis that thrombin and fibrinogen combine in an enzyme-substrate type of complex prior to the formation of fibrin.

Blood volume changes in men exposed to hot environmental conditions for a few hours. NATHANIEL GLICKMAN (by invitation), M. M. MONTGOMERY (by invitation), FORD K. HICK (by invitation) and ROBERT W. KEETON. *Department of Medicine, University of Illinois College of Medicine, Chicago.*

Early reports of studies on blood volume changes occurring in various

[1] Assisted by a grant from the Horace H. Rackham research fund.

environments, using indirect methods (hemoglobin, total solids, etc.) showed somewhat conflicting results. Later studies, using direct methods (dye injection or carbon monoxide) yielded consistent results in experiments of prolonged exposure. Previous work has indicated that plasma volume is not constant from one week to the next. Hence, it was deemed necessary to determine on the same morning the control and experimental plasma volume. The dye, T-1824, was used.

Normal subjects, male medical students, were observed in an air conditioned room. They were at basal condition, nude, and had spent the night in the experimental room under comfortable conditions (28.6°C dry bulb and 19.7°C wet bulb). The second plasma volume was made after a predetermined length of exposure to the desired hot environment (dry bulb 37.2°C or 44.4°C, wet bulb 18.7 to 30.1°C). This compelled the heat loss to be entirely evaporative.

Changes in plasma volume, hemoglobin, hematocrit, total proteins, A/G ratio, red and white blood cells, pulse rate, body temperature and weight loss will be reported.

In 6 experiments (exposure of 59 to 160 min.) there was an increase in circulating plasma volume, red cell mass and serum proteins. In 4 experiments (exposure of 69 to 226 min.) there was a decrease in circulating plasma volume and variable changes in the red cell mass. In 14 experiments (exposure of 60 to 205 min.) there were no significant changes in circulating plasma volume, red cell mass, serum proteins or blood counts.

The data indicate that 1, with increase in plasma volume the new fluids were contributed by blood from the body reservoirs; 2, with decrease in plasma volume water was lost by evaporation from the blood plasma; and 3, with no change a summation of the two adjustment factors described above neutralized each other. Thus a considerable quantity of fluid can be requisitioned from the tissues and evaporated from the blood plasma without affecting the circulating plasma volume.

The non-coagulability of menstrual fluid. HELEN I. GLUECK (by invitation) and I. ARTHUR MIRSKY. *The May Institute for Medical Research, The Jewish Hospital, Cincinnati, O.*

Much speculation still exists concerning the factors responsible for the non-coagulability of menstrual fluid. This report deals with an investigation of this problem by means of more modern methods. The blood was collected from normal female subjects by means of a Ramses pessary inserted during the night and removed in the morning. From 2 ml. to 10 ml. of menstrual fluid was thus collected in from 8 to 10 hours.

Coagulation did not occur when the menstrual fluid was mixed with an equal volume of a preparation of prothrombin free fibrinogen (Smith, Warner and Brinkhaus. J. Exper. Med. **66:** 801, 1937) or when mixed with an equal volume of activated serum thrombin preparation (Smith, Warner and Brinkhaus). However, when the menstrual fluid was mixed with the thrombin preparation plus fibrinogen, coagulation occurred within the same interval of time as did the control which consisted of a mixture of equal volumes of saline, thrombin, and fibrinogen.

When menstrual fluid was mixed with equal volumes of calcium chloride (0.28 per cent) thromboplastic substance (Quick. Science **92:** 113, 1940) and fibrinogen, coagulation did not occur.

The addition of menstrual fluid to a solution of fibrinogen and thrombin did not prevent the coagulation of the fibrinogen.

The above studies suggest that normal menstrual fluid does not contain prothrombin, thrombin, fibrinogen which can be activated, or anticoagulants. The mechanism responsible for the absence of these factors in the menstrual fluid will be discussed.

Thyroid extract and dinitrophenol on intestinal motility. N. M. GLYER and M. J. OPPENHEIMER (introduced by D. A. Collins). *Department of Physiology, Temple University, School of Medicine, Philadelphia, Pa.* (Read by title.)

A study was made of the effects of thyroid extract and alpha 1,2,4, dinitrophenol sodium upon the rate of intestinal contractions. The test objects were trained dogs with intestinal loops exteriorized at various levels, with nerve and blood supply intact, and unstimulated by distention.

Thyroid extract (0.4–0.6 gm/kgm.) was administered by mouth in single daily doses for 14–50 days. Dinitrophenol (0.015 gm/kgm.) in single subcutaneous doses was studied in acute experiments.

Thyroid extract in doses which have been shown to elevate metabolism fifty percent, does not increase the rate of contraction except in a loop placed just above the ileocecal valve. This was the lowest of eight loops studied. Dinitrophenol in doses which have been shown to elevate metabolism two hundred to five hundred percent, does not increase the rate of contraction.

The propagation and distribution characteristics of the action potential in the frog's semi-membranosus, sartorius, and biceps muscle.[1] HAROLD GOLDBERG (by invitation) and J. A. E. EYSTER. *Department of Physiology, University of Wisconsin, Madison.*

A study of the frog's gastrocnemius muscle excited through its motor nerve previously reported showed that the axial action potential distribution along the surface is polar and not simply propagated. The potential distribution is non-propagated during the first $\frac{1}{5}$ and last $\frac{2}{5}$ of the action potential time duration and propagated only during the intervening interval.

The same techniques, the method of instantaneous field distribution determinations and the method of simultaneous recording of unipolar and differential potential time curves, have been applied to the semimembranosus muscle excited through its motor nerve. The distribution in this case is also polar and shows the same relative intervals of propagation and non-propagation found in the gastrocnemius. The distribution at any instant, however, is more complex than that for the gastrocnemius.

The method of simultaneous unipolar and differential potential time curves, which yields information as to the propagation characteristics and polar nature of the distribution without revealing individual details has also been applied to the sartorius and biceps muscles excited through their motor nerves. These too show a polar distribution whose propagated intervals again give the same distribution as those in the gastrocnemius.

The results of this study for four muscles of the frog, each excited through their motor nerve, the gastrocnemius, semi-membranosus, sar-

[1] Supported in part by a grant from the Wisconsin Alumni Research Foundation.

torius, and biceps, shows that the axial action potential distribution along the surface in each is polar, is non-propagated during the first $\frac{1}{5}$ and last $\frac{2}{5}$ of the action potential duration, and is propagated in the intervening period.

Forced intestinal drainage as a method of extrarenal elimination of urea. ARNOLDUS GOUDSMIT. *Gastrointestinal Section of the Medical Clinic, University Hospital, and the Department of Physiology, The School of Medicine, University of Pennsylvania, Philadelphia.* (Read by title.)

A modified small intestinal tube of the Miller-Abbott type with a balloon communicating with one lumen, was passed through the stomach and duodenum, and the tip was allowed to proceed well into the upper jejunum. The balloon placed immediately oral to the tip, was then inflated. A hypertonic solution of sodium sulfate was introduced at regular intervals directly into the duodenum by means of an additional lumen. To secure the intestinal contents suction was applied immediately proximal to the balloon. Additional means were provided for introducing fluid beyond the tip, to avoid dehydration. The volume of the aspirated fluid and its content of ammonia, urea, chloride and sulfate were determined.

Three normal human volunteers and two patients with far advanced renal insufficiency were studied. The average volume of the fluid removed amounted to 655 and 399 cc. per hour for the normals and the patients, respectively. The average concentration of urea in the drained fluid was 93 and 75 per cent of that in the blood for the normals and the patients respectively. These concentrations compare favorably with those observed in peritoneal lavage fluids. In one patient, in the course of seven hours and forty minutes, 3300 cc. of fluid, containing somewhat more than ten grams of urea, were removed. From the other patient, who had approximately one half as much urea in the blood, more than four grams were removed in seven hours and forty-five minutes. As was expected no significant changes in the concentration of urea in the blood were observed subsequent to these drainages. It is suggested, nevertheless, that a procedure similar to the one described but extended over a longer period of time may be useful in supplementing temporarily the excretory functions of the kidney, in conditions where the organ is the seat of severe disease.

Bromide space, thiocyanate space and the measurement of extracellular fluid volume. ARNOLDUS GOUDSMIT, LAWRENCE LOUIS (by invitation) and JOHN C. SCOTT. *Departments of Physiology, University of Pennsylvania and Hahnemann Medical College, Philadelphia.*

A definite volume of a solution containing a mixture of approximately 31 milliequivalents of sodium bromide and 12 milliequivalents of sodium thiocyanate was injected intravenously into normal male human subjects. At various intervals thereafter blood was drawn and the serum analyzed for bromides and thiocyanates. A control sample of blood taken before the injection was subjected to the same procedures. Loss of bromides and thiocyanates with the urine was determined. The difference of the amount injected and the amount excreted divided by the concentration per kilogram of serum water was calculated to indicate bromide space and thiocyanate space, respectively, at a given time.

The control samples always contained materials behaving as bromide and as thiocyanate. Failure to take this into consideration might have changed the values calculated as bromide space as much as eight per cent and the thiocyanate space as much as seventeen per cent. Thiocyanates usually are present in higher concentration in the serum of smokers than in the serum of non-smokers.

Bromide space invariably is larger than thiocyanate space. Taking the values corresponding to ninety minutes after the injection, the excess in seven experiments amounted to an average of 1.90 kilogram (s.d. ± 0.43) or 11.2 per cent (s.d. ± 2.5) of the bromide space. Both bromide space and thiocyanate space are minimal shortly after the injection. Their values increase steadily so that between 90 minutes and 400 minutes after the injection they have increased an average of 1.75 and 0.88 kgm., 10.5 (s.d. ± 1.3) and 6.8 (s.d. ± 3.6) per cent of their values at 90 minutes, for bromides and thiocyanates respectively. Nevertheless in the majority of instances no further increase is noted beyond eight to sixteen hours after the injection indicating that at such times bromides and thiocyanates have reached their final distribution. These final volumes might well prove to represent spaces of definite biological significance.

The support of the John and Mary Markle Foundation is gratefully acknowledged.

The oxidation of estrogens by phenolases. MARK GRAUBARD[1] and GREGORY PINCUS. *The Physiological Laboratories, Clark University, Worcester, Mass.*

A phenolase characterized as laccase prepared from the mushroom Russula delica was found to oxidize the estrogenic substances estriol, estradiol and estrone. These latter are extremely insoluble in water but were made available for enzymic action by having the mixture in a 26.66 per cent concentration of alcohol. This concentration does not inactivate the enzyme. Oxygen uptake is measured in Warburg respirometers. Whenever an uptake of oxygen is observed a precipitate appears in the system. Addition of boiled enzyme shows no oxygen absorption and leads to no formation of a precipitate. By the same methods laccase was found to have no action on dehydroandrosterone, testosterone and progesterone.

Since mushroom tyrosinases are often mixed with laccase, potato tyrosinase was used with the above estrogenic and androgenic substrates and was found not to act on any of them.

The action of the cytochrome oxidase-cytochrome system was tried upon the same substrates because of the general resemblance this system bears to laccase. It was found that the minimum amount of alcohol necessary to keep the substrates in solution was sufficient to inactivate the oxidase. The substrates were therefore suspended in lecithin under which circumstances they are acted upon by laccase. The cytochrome oxidase system was found under these conditions not to act upon any estrogens or androgens.

[1] Fellow of Finney-Howell Research Foundation.

The effect of inorganic ions on gastric secretion in vitro. JOHN S. GRAY and J. L. ADKISON (by invitation). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

We have previously shown that the isolated gastric mucosa of the frog will secrete acid when properly mounted in a bath consisting of two chambers separated by the gastric membrane (Am. J. Physiol. **130:** 327, 1940). The effect of changing the concentration of inorganic ions in the nutrient solution on this *in vitro* secretion has been investigated. In eleven control experiments the pH of the secretory solution fell to an average level of 2.9 in five hours. When the calcium ion concentration was doubled, or the ion omitted entirely, the pH fell more slowly, did not fall as far, and began to rise again after several hours. Doubling or removing the potassium ion had similar but less extensive effects. Alteration in either direction of the Mg ion concentration resulted in mild inhibition, consisting mainly in delaying the onset of secretion. The omission of PO_4 or HCO_3 ions had the least effect, in fact the lowest pH so far recorded, namely, 1.49, occurred in the absence of HCO_3 ion.

Colon activity in the intact animal. STEPHEN W. GRAY (introduced by F. R. Steggerda). *Department of Physiology, University of Illinois, Urbana.* (Motion picture demonstration.)

By synchronizing the x-ray tube and shutter action of a motion picture camera, pictures of colonic activity and defecation were taken from the fluoroscopic screen at a rate of 3 per second. In all cases the colon had previously been made opaque to x-rays with barium enemas or thorotrast injections into the walls of the colon, and the animals used were unanesthetized.

The equipment used allowed continuous pictures to be taken for periods up to 15 minutes, at 88 to 98 kilovolts and 10 to 20 milliamperes. The actual calculated amount of radiation given the animal was far less than the estimated erythema dose.

Studies on the colon of the intact animal. STEPHEN W. GRAY (by invitation) and F. R. STEGGERDA. *Department of Physiology, University of Illinois, Urbana.*

Cine-radiographs of colonic activity in the intact unanesthetized dog and cat were taken at the rate of 3 per second. The colons were visualized by either injecting thorotrast into the walls of the colon or by the giving of barium enemas.

Under the conditions of the experiment the most common movements of the colon were antiperistaltic waves. In the cat these are confined to the transverse and ascending portions of the colon, while in the dog, the middle segment is the most active. The waves occur at a rate of five to seven per minute. Antiperistalsis is not dependent upon the quiescence of the animal.

The act of defecation has been observed in both the cat and the dog with either of two types of movements being responsible for the passage of the semi-liquid enema material, one originating in the lower colon and the other in the upper portion. The latter type is produced by a tonic contraction of the whole upper colon with a longitudinal shortening of the entire bowel. Before the expulsion of feces has entirely ceased strong

antiperistaltic movements, from six to ten per minute, commence in the middle of the colon and refill the upper part in spite of rather definite tonic contraction. All during the time the observations were made, the lower colon remained surprisingly inactive.

The influence of sex on hepatic glycogen formation and destruction. ISABELLE GRAYMAN (by invitation), NORTON NELSON (by invitation) and I. ARTHUR MIRSKY. *The May Institute for Medical Research, The Jewish Hospital, Cincinnati, O.*

Deuel and his co-workers (J. Biol. Chem. **104:** 519, 1934) have observed that fasting results in a more rapid depletion of the liver glycogen of the female than that of the male. In view of the importance of this phenomenon, their studies were repeated using a slightly different approach.

After a preliminary fast of 18 hours, male and female rats were given a subcutaneous injection of 200 mgm. glucose per 100 gram of body weight. The blood sugar and liver glycogen were determined in different animals at various intervals for six hours after the injection of glucose. In that manner the rate of formation and destruction of liver glycogen was obtained.

The blood sugar curve revealed a more rapid return to the fasting initial level in the female rats than in the male rats. At the same time, the curve of hepatic glycogen revealed a lower peak and a more rapid return towards the fasting level in the female than in the male. These observations are in accord with those of Deuel et al. and suggest that an increased demand for carbohydrate by the extrahepatic tissues occurs in the female. The significance of this data will be discussed.

Diabetic herbivora. PAUL O. GREELEY. *Department of Physiology, School of Medicine, University of Southern California, Los Angeles.*

The depancreatized rabbit and goat have been studied with regard to blood sugar levels, glycosuria, ketonuria, survival without insulin, weight changes with and without insulin, and fasting nitrogen excretion.

Both of these animals were able to survive for a considerable time without insulin. Both had elevated blood sugars when fed, the rabbit's being on the average about 400 mgm. per cent, and that of the goat about 220 mgm. per cent. After several days of fasting, the blood sugars fell to normal levels. Ketonuria was constantly present in the goat but rarely observed in the rabbit. Glycosuria attains a maximum of about 50 gm./24 hrs. in each animal. In order to gain weight and approach a normal state insulin was required. The fasting nitrogen excretion was apparently about the same as found for the fasting normal animal.

Zero blood pressure level.[1] HAROLD D. GREEN. *Department of Physiology, Western Reserve University Medical School, Cleveland, O.*

In many problems the value of quantitative venous, intraventricular, or atrial pressure curves from various regions of the conducting system depends on the accuracy with which zero pressure is determined. If, in such experiments on man or animals with closed chests, the center of the ventricle is taken as the zero pressure level, an error is incurred due to the

[1] Aided by a grant from the Commonwealth Fund.

effect of the difference between atmospheric pressure and the intrathoracic pressure surrounding the central vascular structures. It is suggested, therefore, that the zero reference plane be placed at a distance vertically below the center of the ventricle equal to the negative intrathoracic pressure in centimeters of blood.

When optically recorded pressure records are used for the purpose of determining quantitatively cardiac work, peripheral resistance, ventricular filling pressure, etc., it is preferable to record simultaneously the pressure proximal to and that distal to the portion of the vascular bed being studied and then to measure the difference. It has been the practice of some investigators in making this type of study to take each cannula tip as the zero level of pressure for its manometer regardless of differences in level. This arrangement requires use of correction terms to account for the differences in zero level of the manometers, i.e., the Z terms in the Bernouille equation $P_1 + V_1^2/2g + Z_1 + H = P_2 + V_2^2/2g + Z_2 + F$. If, however, one uses the same zero plane for all manometers, then the Z terms reduce to zero and the recorded pressures (P terms) automatically take into account differences in level of the cannulae. The equation then states that the difference in pressure head plus the difference in velocity head between the two points equals the energy added by the heart minus the friction loss $(P_2 - P_1) + \left(\frac{V_2^2 - V_1^2}{2g}\right) - H - F$. It is recommended in this type of experiment, therefore, that regardless of differences in the height of the various cannula tips, the optical manometers be calibrated simultaneously from a mercury manometer, the zero level of which is set as suggested above.

Vitamin A and other fluorescent substances in the retina. RUVEN GREENBERG and HANS POPPER (introduced by F. T. Jung). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, and the Cook County Graduate School of Medicine, Chicago, Ill.*

Thin frozen sections of the albino rat eye were mounted wet and examined in ultraviolet light for their vitamin A content (bright green fluorescence which fades within one-half minute of irradiation) according to the method of Querner (Klin. Wchnschr. **2:** 1213, 1935) and of Popper (Proc. Soc. Exper. Biol. and Med. **43:** 133, 1940).

The preliminary report of von Jancsó and von Jancsó (Biochem. Ztschr. **287:** 289, 1936) showed that vitamin A is present in the light adapted retina and absent from the dark adapted retina, consistent with Wald's formulation of the visual cycle.

We have confirmed and extended these results. Vitamin A is distribu in the eye in two ways: 1, in the ciliary processes, its presence depends on the nutritional status, and is independent of the state of light adaptation; and 2, in the pigment epithelium and in the rod and cone layer the presence of vitamin A does not depend on the nutritional state, but on the state of light adaptation. Within the limits of the method there may be some reduction in amount in vitamin A deficiency.

In the ciliary processes the vitamin A is present in droplets in the capillary endothelial cells and in the fixed connective tissue cells. In the pigment epithelium the vitamin A is present in small droplets and in thin

filaments connecting the droplets, arranged at the corners of the hexagonally shaped cell. The pigment epithelium contains most vitamin A; the rod and cone layer only traces.

In addition the eye showed two other types of fluorescence, not due to vitamin A, which varied with the state of light adaptation: 1, in the dark adapted eye a homogeneous, rust-colored, non-fading, background fluorescence is seen in the pigment epithelium, the rod and cone layer, and the outer molecular layer; 2, in the light adapted eye, this is replaced by a yellow, homogeneous, non-fading, fluorescence. The basis of this change is not yet known.

The diabetes of depancreatized dogs made more severe by administration of foreign proteins, bacteria and locally irritating substances. JAMES A. GREENE and ANN DAVID (by invitation). *State University of Iowa, College of Medicine, Iowa City.*

It has been generally accepted that, during an infection or following the injection of a foreign protein, human diabetes becomes more severe because of a decrease in insulin secretion. If this be true, the diabetes of depancreatized dogs should not become worse under similar conditions. Depancreatized dogs whose diabetes had become stabilized were given injections of typhoid vaccine, skimmed milk, cultures of living bacteria, and turpentine. There were 23 observations upon 7 dogs. The periods of observation varied from 12 to 66 days. The diabetes was not altered in 7 experiments. It was made only slightly more severe in 5, moderately more severe in 10, and extremely severe in 1; 1 dog developed an extreme exacerbation spontaneously. Lipocaic deficiency did not alter this reaction in 1 dog, nor did the presence of small portions of pancreas in 2 dogs. The liver of 1 dog was filled with acacia without altering the reaction.

These observations demonstrate that the diabetes of depancreatized dogs can be made more severe, and render untenable the theory that decreased insulin secretion is the cause of an increase in the severity of human diabetes during an infection or after the injection of a foreign protein. The mechanism of this is not clear. Lipocaic deficiency is apparently not a factor.

Postnatal treatment of rats with sex hormones: the permanent effects on the ovary.[1] R. R. GREENE and M. W. BURRILL (introduced by A. C. Ivy). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Female rats were given daily treatment with chorionic gonadotropin, progesterone or testosterone propionate during the first 2 to 4 weeks of life. Others were given single large doses of testosterone propionate or estradiol dipropionate at birth. Sixty-three of these animals were killed 3 to 18 months later. The ovaries in each treated group were uniformly small and no recent or old corpora lutea were present. Markedly enlarged oviducts showing inflammatory changes were found in animals of the chorionic gonadotropin, androgen and estrogen groups. Vaginal cornification and squamous metaplasia of the uterus were found in some

[1] Aided in part by a grant from the Josiah Macy Jr. Foundation.

animals of all the groups. This latter finding is indicative of constant estrogenic stimulation. In agreement with this, constant estrus smears were obtained in animals of the estrogen and gonadotropin group which were smeared daily for several months. However, the smears became diestral in type after administration of large doses of chorionic gonadotropin and, at autopsy, large corpora lutea were found in the ovaries.

It is apparent from this latter finding that the primary disturbance in these animals is in the anterior pituitary and not in the ovary. This alteration in pituitary gonadotropic function cannot, however, be considered as due to a "masculinization" of this gland as has been claimed. It seems that the administration or forced production of large amounts of any of the sex sterols in the very young female rat is capable of permanently altering pituitary gonadotropic function.

Secretinase.[1] HARRY GREENGARD, IRVING F. STEIN, JR. (by invitation) and A. C. IVY. *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

The typical response of the pancreas to a single injection of secretin is an initial rapid secretion which gradually diminishes and finally ceases, indicating a disappearance of the hormone from the circulation. This may occur as a result of destruction, excretion, or storage of secretin. The possibility of its destruction in the blood stream was investigated by incubating a standard amount with samples of blood and noting the response of the pancreas to such mixtures with reference to that elicited by control secretin injections. It was observed that such incubation inactivated the secretin to an extent depending on the time in incubation. Inactivation was obtained with dogs' whole blood, plasma, or serum, and not with washed cells. The serum or plasma were deprived of their secretin-inactivating power by heating to 60° for half an hour. The temperature of incubation definitely affected the rate of secretin inactivation, and the optimal temperature was 35–40°C. The rate of inactivation also varied directly with the quantity of serum used. While the extent of inactivation varied somewhat in individual blood samples, it was in all cases progressive with time and complete in 4 to 12 hours. There was no inactivation outside the pH range of 5 to 9; the optimum reaction was at pH 7.0–7.5. All these findings indicate that the mechanism of secretin inactivation is an enzymic one, and it is concluded that the blood serum of dogs contains a secretinase. This principle has also been demonstrated in human blood and in beef blood.

Some biological properties of metakentrin and thylakentrin. R. O. GREEP, H. B. VAN DYKE and BACON F. CHOW (by invitation). *Division of Pharmacology, Squibb Institute for Medical Research, New Brunswick, N. J.*

Using chemically pure metakentrin (ICSH) and biologically pure thylakentrin (FSH) extracted from hog pituitary glands we have reinvestigated many previous studies that had been made with preparations, the purity of which had not been as fully ascertained. While much of our data is confirmatory in nature, a few striking departures from reported

[1] Aided in part by a grant from the Josiah Macy Jr. Foundation.

findings have been encountered. The data all demonstrate that the principle of duality, as applied to extractable pituitary gonadotrophins by Fevold and Hisaw, appears to be established.

Metakentrin stimulated the gonadal interstitial cells of hypophysectomized male and female rats and normal male pigeons. This stimulus evoked secretory activity only in the male rats and this reaction, measured by the response of the anterior prostate gland, was shown to be a specific reaction to this pure gonadotrophin. Upon this basis a method of assaying metakentrin quantitatively has been evolved.

Thylakentrin produces a more selective histological change in the testes than does metakentrin since the reaction is not complicated by secondary androgen stimulation. A satisfactory assay of thylakentrin has been carried out depending upon the weight increase of the testes of hypophysectomized immature rats.

Thylakentrin in high dosage of protein over 10 days caused development of graafian follicles without estrogen secretion (histology of uterus and vagina, uterine weight) or luteinization in hypophysectomized immature female rats; if metakentrin was administered the last 5 days, estrogen secretion could be detected within 48 hours and rapidly progressed to complete estrogenic development of the uterus and vagina accompanied by luteinization and some augmentation of ovarian weight. Metakentrin alone in high dosage over 10 days caused only stimulation of interstitial cells.

The previously reported ability of the luteinizing fraction (Greep. Endocrinology **23**: 154), acting alone to destroy the corpora lutea persisting in hypophysectomized adult female rats, has not been confirmed in limited tests with metakentrin. However, the functional capacity of the corpora lutea of pseudopregnancy in rats was not maintained by metakentrin following removal of the pituitary, as determined by the absence of a decidual response.

The ability of metakentrin to antagonize the action of mare serum gonadotrophin was also investigated.

Observations on the accuracy of the thermostromuhr.[1] D. E. Gregg, W. H. Pritchard (by invitation), R. W. Eckstein (by invitation), T. W. Steege (by invitation) and J. T. Wearn. *Department of Medicine, Western Reserve University, Cleveland, O.*

The direct current thermostromuhr of Baldes was constructed using improved lacquers and lead wires thereby largely eliminating shorts, and breakage of lead wires in chronic experiments. The units were calibrated with an artificial circulation system and rotameter and then applied to coronary arteries of normal dogs for two to six weeks. In some the galvanometer readings were reasonably constant. Then the units were recalibrated either in situ or on the same artery removed to the schema.

Each calibration curve can be duplicated within 10 per cent, but curves obtained at the end of an experiment generally differ widely from the initial curve. Similarly, curves differ considerably on the same unit applied to various vessels in different dogs or in a schema. Accordingly, effects of many physiological variables on unit accuracy were tested in a schema and in dogs.

Variations in blood temperature, viscosity, heart rate, stroke volume,

[1] Aided by a grant from the Commonwealth Fund.

pattern of flow curve, and composition of fluid surrounding unit and artery have minor effects on unit accuracy providing these variables are within physiological limits and that no zero or back flow occurs.

However, extension or shortening of the artery (in acute or chronic experiments), the presence of a small systolic back flow, or zero flow, changes in the elasticity of the pump system, changes in blood flow through surrounding tissues, and relationship of such tissue to the unit, all grossly effect the accuracy of the thermostromuhr (up to 300 per cent). Furthermore, long application of the unit to arteries may narrow the lumen sufficiently to reduce the flow.

Therefore, any interpretation of galvanometer deflection in terms of absolute blood flow is open to grave question.

The effect of sodium sulfapyridine and sodium sulfathiazole on blood sugar and liver glycogen.[1] ESTHER M. GREISHEIMER, ROBERTA HAFKESBRING and HULDA MAGALHAES (by invitation). *Department of Physiology, Woman's Medical College of Pennsylvania, Philadelphia.*

The method used was described by us previously (Greisheimer, Hafkesbring and Magalhaes—Am. J. Physiol. **129:** P 371, 1940).

The following values for blood sugar and liver glycogen were found:

	BLOOD SUGAR	LIVER GLYCOGEN
	mgm. per 100 cc.	*per cent*
Fasting	85	0.25
Control (glucose, no drug)	93	1.48
Fasting; sodium sulfapyridine 10%	134	0.05
Glucose and sodium sulfapyridine 10%	202	0.14
Glucose and sodium sulfapyridine 7.5%	190	0.73
Fasting and sodium sulfathiazole 7.5%	123	0.24
Glucose and sodium sulfathiazole 7.5%	225	1.56
Glucose and sodium sulfathiazole 10%	233	0.80

Sugar determinations on the filtrate before and after fermentation with yeast ruled out the possibility of reduction by drugs.

The administration of a single dose of sodium sulfapyridine is followed by an increase in blood sugar and a decrease in liver glycogen.

The administration of sodium sulfathiazole is followed by a marked increase in blood sugar; in this respect it resembles sodium sulfapyridine. However, in equally toxic doses, it has little effect on liver glycogen. When given in larger doses (10 per cent), in which there is about 50 per cent mortality, it leads to some decrease in liver glycogen. It seems safe to conclude that in equally toxic doses, sodium sulfapyridine is more harmful to the liver with respect to its ability to form and store glycogen than sodium sulfathiazole.

Total and subtotal sympathectomy in man, effect on blood pressure in hypertension. KEITH S. GRIMSON (by invitation), ALF S. ALVING and WRIGHT ADAMS (by invitation). *Departments of Surgery and Medicine, University of Chicago, Chicago, Ill.*

[1] Supported by a grant from the Committee on Therapeutic Research, Council on Pharmacy and Chemistry, American Medical Association.

Total sympathectomy was first demonstrated to be compatible with life in cats and dogs by W. B. Cannon and his associates. This procedure has recently been employed in many studies of experimental renal and experimental neurogenic hypertension. It was found to have a marked blood pressure lowering effect only in experimental neurogenic hypertension.

A three stage surgical technique has been devised for removal of the thoraco-lumbar sympathetic chains, the splanchnic nerves, and the celiac ganglia in man. It has been employed upon eleven patients of the idiopathic or essential hypertension group. Two patients received all three stages. Seven received the two thoracic operations including removal of all of the thoracic and the first lumbar sympathetic chain ganglia together with all of the splanchnic nerves and the celiac ganglia. The remaining two received the abdominal operation including bilateral lumbar sympathectomy and splanchnic section.

Total sympathectomy has been demonstrated to be compatible with a relatively normal existence in man. Postural hypotension has been noted. The heart rate has been decreased. Gastrointestinal, urinary and respiratory functions have not been conspicuously altered. The loss of sweating has given only slight inconvenience. The subtotal two-stage thoraco-lumbar sympathectomy, splanchnic resection, and celiac ganglionectomy have given similar results except that compensatory excessive perspiration about the legs has occurred in warm weather. Blood pressure lowering to relatively normal values has been observed in four of the nine hypertensive patients receiving total or subtotal sympathectomy. Three others have had definite blood pressure lowering but have not reached the normal blood pressure range. One patient had only slight blood pressure lowering and one patient operated upon in a stage of advanced cerebral damage died. These nine patients have been observed from two to ten months. In some instances a tendency toward blood pressure recovery has been noted. Some patchy restoration of sweating has also developed.

Bilateral lumbar sympathectomy and splanchnic section have effected only slight alteration of the blood pressure.

Does the posterior pituitary exert an influence on gastric secretion?

E. G. Gross, W. R. Ingram and N. W. Fugo (by invitation). *Departments of Pharmacology and Anatomy, State University of Iowa, Iowa City.* (Read by title.)

Studies of gastric secretion in patients with diabetes insipidus have been reported which indicate an influence of the posterior pituitary gland, especially in the control of acidity. These observations were made by taking frequent gastric samples. It was believed that these observations should be checked in dogs with Pavlov pouches.

The effect of food and histamine has been studied in these animals before and after section of the pituitary stalk. Typical diabetes insipidus developed after stalk section; the urine volume averaging about 2000 cc. per day. Normal gastric secretion was collected for thirty minutes before the stimulus was given and then collected for four thirty minute periods following the stimulus. Several runs were made before and after section. The volume, pH, total acidity, and free acidity were determined.

Using 200 grams of Pard dog food plus 150 cc. of water as the stimulus, no significant change in the pH, total acidity, and free acidity before and

after section was observed. The average volume of gastric secretion was about 25 per cent less in the periods after section.

When histamine was used as the stimulus, again the only obvious difference in the two periods was the decrease in volume of gastric juice. It may be possible that the decreased volume after stalk section is due to a partial dehydration that may occur in these animals during the two and one-half hour period of collection.

The augmentation of the motor root reflex discharge in the cooled spinal cord of the cat. HARRY GRUNDFEST. *Department of the Laboratories of the Rockefeller Institute for Medical Research, New York City.*

In cats under dial anesthesia and in decerebrate-spinal preparations, the lumbar and sacral regions of the spinal cord were widely exposed. Lead electrodes placed on the dorsal and ventral roots of one segment recorded both the motor and the dorsal root reflexes evoked in response to stimulation of the dorsal root. The exposed cord was flooded with liquid paraffin and its temperature varied by warming or cooling the oil.

Under these experimental conditions, as the temperature was lowered from 38°C. to 30°C., the reflex discharge recorded from the ipsilateral ventral root was progressively augmented and at times components of the potential were increased to four times their height in the warm cord. Records from the corresponding dorsal root showed that the dorsal root discharge was also increased as described by Barron and Matthews and confirmed by Toennies. Thus the discharges from the two roots have similar sensitivities to temperature.

As the cord is cooled, reflexes not obtainable at normal temperatures may appear. In the decerebrate-spinal preparations above described, at normal temperatures no visible ventral root discharge was produced by a single shock to the contralateral dorsal root; while a clearly observable discharge was evident when the cord was cooled below 36°C. At 30°C. the discharge was well defined.

Carbohydrate relationships in the rat. M. MASON GUEST (by invitation), E. L. SCOTT and J. J. MCBRIDE (by invitation). *Department of Physiology, College of Physicians and Surgeons, Columbia University, New York City.*

The coefficient of dispersion for rat liver and muscle glycogen concentrations has been reduced to 10 per cent by the standardization of the technical procedures and the biological material. Liver glycogen concentrations may be predetermined at any desired level between approximately zero and eight per cent of the tissue wet weight to a precision sufficient for most experimental purposes by controlling the nutritional condition of the experimental animal.

Analyses of the compiled data by regression methods have indicated several physiological relationships. Thus the log. value of the percentage of liver glycogen in fasted rats varies directly with the blood sugar concentration. In contrast, rats killed at the end of the feeding period exhibit little variation in the mean blood sugar concentration regardless of the liver glycogen level. Similarly a positive correlation has been shown between the muscle and liver glycogen concentrations for both fed and fasted rats.

The plot of muscle glycogen against the blood sugar concentration of

fasted rats reveals a suggestive distribution of the individual points. Little variation in the muscle glycogen concentration occurs in a blood sugar range between 50 and 100 mgm. per cent, but above this level, although there is a much greater variation of the individual points, the general tendency is toward glycogen storage. In the fed groups there is little evidence of correlation between blood sugar and muscle glycogen concentrations.

The neuropathology of vitamin E-deficiency in the rat. W. DE GUTIÉRREZ-MAHONEY, KARL E. MASON and HOMER SWANSON (by invitation). *Laboratory for Neuropathology, Vanderbilt University School of Medicine, Nashville, Tenn.*

The entire nervous system of baby and adult rats, born of E-deficient mothers and maintained on E-deficient diets, were examined by the following methods: Nissl for cells; Herxheimer, Kulschitzky, Marchi, Spielmeyer and 1 per cent Osmic acid for myelin sheaths; Gros-Bielschowsky and Ramon y Cajal for axis cylinders. The animals were sacrificed for histological study at various ages from 18 to 450 days.

The changes in the baby and adult rats were similar on the whole. The nerve cells were generally seriously altered so that at first the cytoplasm was hyperchromatic, the Nissl pattern was lost, and neither nucleus nor nucleolus were visible. The cell bodies became thin, long and sharply outlined without accompanying phagocytic cells. A later stage was characterized by diminution in the stainable cytoplasm with vacuolation, and scalloping of the cell borders with attendant phagocytes, finally leaving merely the ghosts of the original cells. The dendrites became stained for long distances and took on bizarre shapes. These changes affected almost all groups of cells, but were most striking in cells of the anterior and posterior horns of the spinal cord, of the cranial nerve nuclei, of the olives, and of the pontine nuclei. The Purkinje cells of the cerebellum and the large and small cells of the cortex were also involved.

The alterations did not resemble the retrograde reaction of Nissl nor the ordinary chronic cell change. Some were similar to post mortem changes, and when first seen they were attributed to this, but the disturbances were seen regularly in animals which were killed and the tissues fixed immediately.

Degenerative changes in the white matter were demonstrated by all suitable methods noted above, and were characterized by swelling, fragmentation, ball formation and disappearance of myelin. The peripheral nerves, the posterior tracts and the spinocerebellar tracts were severely affected, while the pyramidal pathways and cerebral white matter showed only a moderate degree of disturbance.

The silver stains demonstrated swelling, fragmentation, and dissolution of the axis cylinders of the nerves, spinal cord, cerebellum and brain.

The glial reaction was well marked in all areas affected beyond the initial stage.

Chemical studies in traumatic shock. H. GUTMANN (by invitation), WM. H. OLSON (by invitation), H. H. KROLL (by invitation), S. O. LEVINSON (by invitation) and H. NECHELES. *S. Deutsch Serum Center and the Department of Gastro-Intestinal Research, Michael Reese Hospital, Chicago, Ill.*

Chemical changes were studied in the anesthetized dog following trauma applied to the thigh. Hemoglobin, hematocrit values, plasma protein, arterial O_2 and CO_2, arterial lactic acid and plasma potassium were determined simultaneously. The arterio-venous O_2 difference and arterial pH also were measured in a few cases. Carotid blood pressure was recorded with a mercury manometer. Circulation time was estimated by the respiratory response to the intravenous injection of sodium cyanide. Shock was assumed to develop when the blood pressure had fallen to a level of 50 to 60 mm. Hg.

During shock the following changes were found to occur. There is a significant drop of CO_2 and O_2 in the arterial blood concurrent with a steep rise of the lactic acid concentration and a decrease of the pH. The depletion of the alkali reserve cannot be accounted for on the basis of the accumulation of lactic acid alone. The arteriovenous oxygen difference is markedly increased. Specific gravity of whole blood and plasma protein concentration change inversely as a rule. Hemoglobin and hematocrit readings usually are indicative of hemoconcentration. The plasma potassium may be slightly increased as a terminal event. A transient rise in the plasma potassium has also been observed following severe traumatization. There is evidence suggestive of a possible relationship between the degree of tissue damage and the K content of the blood. While blood pressure and circulation time may be restored to normal at least temporarily by the infusion of blood substitutes it appears in our experience thus far, that the chemical changes in traumatic shock are partially but not completely reversed by restoration of circulating volume.

Electrical rectification in the giant axon of the squid. RITA GUTTMAN (introduced by Kenneth S. Cole). *Marine Biological Laboratory, Woods Hole, Mass. and Department of Physiology, College of Physicians and Surgeons, Columbia University, New York City.*

Measurements of the resistance of the squid giant axon have been made by means of a direct current Wheatstone bridge between one injured end of the axon, placed in KCl, and the other end immersed in sea water, with the inter-electrode region hanging in oil. During the passage of small currents through the axon, the resistance of the fiber does not depend upon the magnitude and direction of the current. But when larger currents are used this is no longer true, i.e. if the uninjured end is the anode, the overall resistance of the axon is greater than that found during the passage of small currents while at the cathode the reverse is true. The apparent resistance of the nerve fiber, then, depends upon the magnitude and direction of the current flowing through it. The nerve fiber thus does not obey Ohm's Law and is an electrical rectifier which permits current to pass more easily in one direction than the other, rather than a pure resistance.

It seems probable that the rectifier effect is confined to the membrane and these results confirm the observations of Cole and Curtis, and Cole and Baker (J. Gen. Physiol., March, 1941, in press) obtained with different methods. When both ends of the axon were uninjured, Cole and Hodgkin (J. Gen. Physiol. **22**: 671, 1939) were unable to detect rectification because the increase in resistance at the anode approximately compensated for the decrease at the cathode.

Cocaine and veratrine cause progressive and reversible loss of rectifica-

tion. As the axon dies, excitability is lost, and the rectification and the resting potential disappear. When the fiber is completely dead, there is no rectification and the fiber acts as a pure resistance.

Such a rectification is to be expected if the membrane conductance is a measure of ion permeability and this permeability is increased at a cathode and decreased at an anode. Also, rectification has been suggested as an explanation of some electrotonic and excitation phenomena.

Recovery after sulfonamide drugs.[1] ROBERTA HAFKESBRING and ESTHER M. GREISHEIMER. *Department of Physiology, Woman's Medical College of Pennsylvania, Philadelphia.*

The purpose of these experiments was to study recovery in blood sugar and liver glycogen in rats after sodium sulfapyridine and sodium sulfathiazole. Two series of experiments were performed: one, after a single dose of 7.5 per cent sodium sulfapyridine or sodium sulfathiazole (1 cc. per 100 grams), and the second, after 4 doses of one of the drugs.

The results are as follows:

CONTROLS, GLUCOSE, NO DRUGS	BLOOD SUGAR 93 SODIUM SULFAPYRIDINE		LIVER GLYCOGEN 1.48 SODIUM SULFATHIAZOLE	
	Blood sugar	Liver glycogen	Blood sugar	Liver glycogen
1 dose, 72 hours before exper...	95	1.39	88	1.45
1 dose, 48 hours before exper...	90	1.39	87	1.66
1 dose, 24 hours before exper...	102	0.81	86	1.41
4 doses, 120, 96, 72, 48 hours before experiment.....	108	0.76	95	1.36

Blood sugar and liver glycogen remain at approximately the control level after single doses of sodium sulfapyridine or sodium sulfathiazole, 72 and 48 hours before the experiment. Liver glycogen is still about 50 per cent below the control value 24 hours after the administration of a single dose of sodium sulfapyridine.

After repeated doses on 4 consecutive days, allowing 48 hours for recovery, liver glycogen is unchanged when sodium sulfathiazole has been used, but reduced to approximately one-half its control value when sodium sulfapyridine was used.

The results of this series of experiments show that after four doses of sodium sulfapyridine on consecutive days, the liver is still far below normal in its ability to form and store glycogen, 48 hours after the drug has been discontinued, and even after a single dose of sodium sulfapyridine, does not completely recover within 24 hours. The effect on the liver of similar doses of sodium sulfathiazole is negligible.

The adrenals and the insulin content of the pancreas. R. E. HAIST and H. J. BELL (introduced by C. H. Best). *Department of Physiological Hygiene, University of Toronto, Toronto, Canada.*

Female rats weighing between 110–150 grams were adrenalectomized,

[1] Supported by a grant from the Committee on Therapeutic Research, Council on Pharmacy and Chemistry, American Medical Association.

given 1 per cent sodium chloride solution to drink and fed a balanced ration ad libitum for 3 to 10 days. Control groups of rats with intact adrenals were also given 1 per cent sodium chloride to drink and received the same diet and the same caloric intake as the adrenalectomized animals. No significant difference was found between the insulin content of the pancreas in the adrenalectomized and paired-fed control groups. Adrenalectomized rats without the 1 per cent sodium chloride showed a reduction in the insulin content of the pancreas but this seems to be related to their lower food intake.

Adrenalectomized animals given 1 per cent sodium chloride but fed a diet consisting largely of fat, showed a marked fall in the insulin content of the pancreas, approximately equivalent to that found in the paired-fed control animals fed a similar diet. These experiments demonstrate that the reduction in the insulin content of the pancreas resulting from the feeding of fat can be obtained in the absence of the adrenal glands. Since in a previous experiment it was shown that the effect of fat feeding can still be obtained in the absence of the pituitary gland, it seems probable that the effect of fat feeding on the insulin content of the pancreas results either from a direct influence on the pancreas or because of effects produced in liver or peripheral tissues.

In another experiment rats were fed a balanced diet and given 3.6 to 5.6 cc. of a potent adrenal cortical extract daily by mouth. Control groups received the same diet and the same caloric intake but no cortical extract. There was no significant difference in the insulin content of the pancreas in the groups receiving the extract and those that did not.

The water and fat content of the skin of the albino rat on a high carbohydrate and a high fat diet. JOHN HALDI, GLENVILLE GIDDINGS (by invitation) and WINFREY WYNN (by invitation). *Laboratory of Physiology and the Department of Medicine, Emory University, Georgia.* (Read by title.)

The effect of a high carbohydrate and a high fat diet on the water content of various organs of the albino rat was determined in a previous investigation (Am. J. Physiol. **128:** 537, 1940). In view of the clinical reports that the water content of the skin is affected by diets containing a large amount of carbohydrate or of fat, we have extended our observations and found that the water in the skin was reduced to a marked degree by a high carbohydrate diet and to a greater extent by a high fat diet.

Litter mates, allowed to eat and drink *ad libitum* were fed 70 days on a stock, a high carbohydrate and a high fat ration, and were then decapitated. The animals on the stock ration served as controls. The carbohydrate ration contained 70 per cent sucrose and no fat; the fat ration 50 per cent lard and 20 per cent sucrose. The skin was removed from the same region of the back of all the animals after decapitation, stripped of adherent fat and adjacent portions analyzed for water and fatty acid content.

The average water content of the skin on the stock, carbohydrate and fat rations was, respectively, 60.3, 55.1 and 51.4 per cent of the wet weight in the males and 51.1, 43.7 and 39.2 per cent in the females. The amount of water present was in inverse ratio to the fatty acid content which was 6.6, 9.3 and 17.6 per cent, respectively, in the males, and 12.0, 20.1 and

39.2 per cent in the females. It will be observed that there was a sex difference in the composition of the skin as shown by the percentage water and fat content.

The blood of the males on the three rations contained 78.5, 80.1 and 80.4 per cent water, respectively, and that of the females 78.4, 80.1 and 80.3 per cent. The determinations of water content were made on approximately 5 cc. of blood obtained from the decapitated animal.

In previous experiments it was found that the same high fat ration fed for the same length of time as in the present experiments did not produce ketosis.

Effects of the administration of d-l threonine and d-l allothreonine to the phloridzinized dog.[1] W. KNOWLTON HALL (by invitation) and A. G. EATON. *Departments of Biochemistry and Physiology, Louisiana State University School of Medicine, New Orleans.*

Female dogs were given subcutaneously 1 gram of recrystallized phloridzin in olive oil daily. Control periods were run on the third and fourth days, and thereafter either d-l threonine or d-l allothreonine was administered on alternate days by the intraperitoneal or subcutaneous route. Total nitrogen, glucose, urea nitrogen and amino nitrogen determinations were made on each 24 hours urine sample.

Contrary to our findings in rats d-l allothreonine produced no extra sugar. In every experiment d-l threonine gave extra glucose. A large proportion of both the d-l threonine and the d-l allothreonine was excreted into the urine. As a result the amount metabolized so small that we are unable to state definitely how many carbon atoms of the threonine molecule go to glucose.

Androgen, a prime factor in acne. JAMES B. HAMILTON. *Department of Anatomy, Yale University School of Medicine, New Haven, Conn.*

A series of eunuchoid individuals, that is those who did not mature sexually, never had acne. Upon administration of male hormone substance, testosterone propionate, to eunuchoid and castrate men, to prepubertal boys and to ovariectomized women, some of these individuals acquired comedones, papules and pustules. When the treatment with hormone was terminated, the acneform responses diminished and began to disappear. Additional courses of medication given to these patients at later dates were accompanied by similar conditions with subsequent disappearance of the lesions during intervals between treatment with testosterone compounds. A close relationship of this acne to male hormone substance is attested by the rapidity with which the lesions fade upon cessation of androgenic therapy, and by the fact that a developing papule which has undergone retrogression may again become inflamed if treatment is soon resumed. In appearance the lesions are like those found at puberty. Acne occurred in most but not all patients receiving adequate doses of hormone. Differential susceptibility in the present series does not appear to be determined by color of hair or skin or by the previous occurrence of puberty; men who pass through puberty before orchidectomy may or may not exhibit acne upon treatment. The greatest severity of erup-

[1] Aided by a grant from the Committee on Scientific Research of the American Medical Association.

tions was observed in a man with a history of severe acne as an adolescent. Discussion is given of the levels in the bodily fluids of androgenic, estrogenic and gonadotropic (follicle-stimulating) hormone in patients with and without acne and of therapeutic implications. Sexual hormones may also, perhaps, bear a relation to furuncular folliculitis in adolescence. The fact that male hormone substance may induce acne in susceptible individuals is not proof that the sole cause of acne is the action of male hormone substance. Other factors and an "innate predisposition to acne" (as evidenced by identical twins) are undoubtedly of great inportance. Androgenic substances stimulate also sebaceous secretions in various parts of the body not subject to acne, such as in the preputial gland.

Cardiac and aortic contributions to the human ballistocardiogram. W. F. Hamilton and Philip Dow. *Department of Physiology and Pharmacology, University of Georgia School of Medicine, Augusta.*

The reawakening of interest in the recoils of the heart and great vessels, and more particularly the recent employment of them as an index of cardiac output make advisable a critical study of these forces and an evaluation of the roles played by each of them in producing the resultant movements of the body.

The apparatus to be described differs from that of Starr et al. in that the subject sits upright instead of lying supine. A chair is suspended from a short stiff spring leaf and its vertical movements are translated mechanically, magnified optically, and recorded photographically. This arrangement seems to permit a higher net frequency of vibration in the recording system as a whole than is found in those previously reported. The deflections of the light beam can be calibrated simply by the addition of weights to the chair with the subject in it.

Ballistocardiograms have been taken simultaneously with records of the carotid and femoral pulse. The pulse records permit identification of the standing waves of the aortic windkessel. These standing waves can be shown to quicken when the subject assumes an upright or semierect posture, when the blood pressure rises and when the windkessel is restricted by a tourniquet about the thighs. In all these cases the pulse wave transmission time from the cardiac to the peripheral boundary of the windkessel is shortened and the period of the balistic waves is shortened equally. On the average the I wave is simultaneous with the start of the femoral pulse. The K wave with its peak, the M wave with the postincisural plateau of the carotid and with the femoral dip, and the O wave with the diastolic rise of the femoral pulse. The ballistic waves are a faithful record of the periodic surging of blood in the aortic windkessel.

Acknowledgment is gratefully made to the Macy foundation for a grant which aided this research.

The effect of botulinus toxin on the mortality and time of death of developing chick embryos.[1] Dorothy M. Hamre and Clayton S. White (introduced by R. W. Whitehead). *Departments of Bacteriology and Physiology and Pharmacology, University of Colorado School of Medicine, Denver.*

Through an air-cavity opening in fertile eggs incubated for 14, 16 and 17

[1] Aided by a grant from the Dazian Foundation for Medical Research.

days a concentrated, partially purified type A botulinus toxin was dropped on the chorio-allantoic membrane exposed by removal of the shell membrane. Series I (107 embryos) received 10 minimal lethal doses as tested on 20 gram white mice. Series II (162 embryos) received 100 minimal lethal doses. The eggs were sealed with cellophane and incubation continued. The embryonic time of death was fixed by observations at autopsy.

Percentage mortality in series I was 58.8 for 52 experimental embryos and 30.9 for controls receiving boiled toxin; in series II, 89.7 for 78 intoxicated embryos and 31.0 for controls. The ratio, per cent difference/σ difference, in series I was 2.61, in series II, 7.6.

Of the dead controls in series I and II 58.8 and 57.7 per cent respectively died on or before the 18th day compared to 34.4 and 14.3 per cent for experimental animals. In contrast the percentage of controls dying on or after the 21st day was 17.6 and 19.3 in series I and II compared to 41.5 and 54.3 for experimental groups.

It is concluded that botulinus toxin in sufficient dosage kills chick embryos, but only after the time when the source of the oxygen begins to shift from the chorioallantoic membrane circulation to that of the lungs. Chick embryos die from respiratory failure as do other botulinus intoxicated animals.

The relation of central nervous system depression and stimulation to biochemical changes occurring in cerebral blood. CARROLL A. HANDLEY (by invitation) and H. MORROW SWEENEY. *School of Medical Sciences, University of South Dakota, Vermillion.*

Dogs were prepared for chronic experiments by removing a section of bone over the superior sagittal sinus. Oxygen, carbon dioxide and blood sugar determinations were made on arterial, sinus and popliteal venous blood drawn from these trained animals during the resting state, following the depressant (morphine, sodium pentobarbital) and following the stimulant (amphetamine, metrazol). Respiratory rate and depth, pulse rate, metabolism and cardiac output were also studied.

During morphine depression, the administration of amphetamine usually caused an increased uptake of glucose by the brain, as measured by the glucose difference between arterial and sinus blood, while the oxygen uptake, though in the same general direction was even less constant. These variables are probably due to changes in blood flow induced by the amphetamine. Central stimulation is apparent, however, from respiratory and pulse rate changes and in some cases the animals were awakened from deep morphine depression.

Studies with metrazol following sodium pentobarbital anesthesia show that both the oxygen and glucose uptake by the brain are increased.

Prevention of experimental gastrojejunal ulcer by enterogastrone.[1] A. P. HANDS (by invitation), G. B. FAULEY (by invitation), HARRY GREENGARD, F. W. PRESTON and A. C. IVY. *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Previous reports from this laboratory have demonstrated the action of enterogastrone in the depression of motor and secretory activity of the

[1] Aided by a grant from the Josiah Macy Jr. Foundation.

stomach, and procedures have been described for the production of uniformly potent and non-toxic (dog) enterogastrone concentrates. In view of the anticipated potentialities of such an agent in the treatment of peptic ulcer, a therapeutic trial has been made in a series of dogs prepared by the Mann-Williamson operation. Following recovery from the surgery these animals were given three daily intravenous injections of enterogastrone concentrate to the exclusion of all other medication. Each injection comprised 50 mgm. of material, which amount halves the gastric secretory response in the total pouch dog and abolishes tone and motility in the stomach of the intact dog for 60 minutes (average). No local or general reactions to the intravenously injected material have been noted, and after a period of 20 weeks after operation 9 out of the 10 animals have uniformly manifested good appetite, maintenance or gain in weight, and no abnormalities in the blood picture, save a slight hypochromic anemia. Since these dogs have remained in good condition *without manifesting an ulcer* for an interval definitely longer than that required for the untreated Mann-Williamson dog to evidence characteristic signs and symptoms of gastrojejunal ulcer, it is concluded that enterogastrone is an effective agent in the prevention of development of this condition.

Reflex production of heart block. H. F. Haney, W. B. Youmans, A. J. Lindgren (by invitation) and A. I. Karstens (by invitation). *Department of Physiology, University of Oregon Medical School, Portland.*

A difference in the distribution of the inhibitory influence of the two vagus nerves on the heart of the dog is indicated by the fact that stimulation of the peripheral end of the cut left vagus often results in partial or complete A-V heart block, a response not obtained with the right vagus. Evidence regarding the physiological significance of this difference has been obtained by means of the following experiments in which vagal inhibition is produced reflexly in the unanesthetized animal as a result of a rise of blood pressure.

Dogs were prepared by sympathetic denervation of the heart and unilateral vagotomy high in the neck. After a suitable period of training, electrocardiograms were recorded before and during appropriate periods after the injection of the pressor compounds, neosynephrin (intravenously) and pitressin (intramuscularly). In the case of neosynephrin a dose was chosen which produced little or no acceleration of the denervated heart.

In a series of 44 experiments on 12 dogs in which the left vagus remained intact the injection of one of the pressor compounds was followed by pronounced bradycardia in all but one case. In 26 of the experiments which included 10 of the animals the injection was followed by either partial or complete A-V heart block and in most cases a prolongation of the P-R intervals.

In a series of 26 experiments on 9 dogs in which the right vagus remained intact the records taken following the injecion of the pressor compounds showed sinus bradycardia in all cases, A-V nodal rhythm in 5 experiments on 3 dogs, and no cases of A-V heart block.

It seems clear that a rise of blood pressure is capable of initiating reflex depression of the S-A node through either vagus and in addition a reflex depression of the A-V node through the left vagus of sufficient degree to produce A-V heart block.

Studies on pain: observations on the hyperalgesia associated with referred pain. J. D. HARDY, H. GOODELL (by invitation) and H. G. WOLFF. *Russell Sage Institute of Pathology in affiliation with the New York Hospital and Departments of Medicine and Psychiatry, Cornell University Medical College, New York City.* (Read by title.)

The so-called hyperalgesia associated with referred pain was investigated to determine whether the over-reaction to pin prick in the hyperalgesic area was due to lowered pain threshold at this site. In a group of 7 patients with diaphragmatic irritation, duodenal distention, or a tooth abscess, it was possible to measure the pain threshold in circumscribed areas of referred pain with hyperalgesia. It was shown by the use of the pain threshold measuring device previously described (J. Clin. Ivest. **19:** 649, 1940), that the pain threshold in the areas specified as hyperalgesic was normal and differed in no way from that of the opposite corresponding area similarly tested. It is inferred that such changes in sensation as occur in hyperalgesia with referred pain are not the result of lowered perception, but rather a change in the reaction to the afferent impulses initiated in the periphery at the usual threshold. Thus these areas are not truly hyperalgesic, as is the case in "sunburn."

The effect of withdrawal of drinking water upon the antidiuretic potency of the posterior lobe of the rat. KENDRICK HARE, ROBERT C. HICKEY (by invitation) and RUTH STILLMAN HARE (by invitation). *Department of Anatomy, Cornell University Medical College, New York City.*

Rats of the same age and sex were divided into groups of seven; one group was kept on the normal diet of dried food with free access to water, and a second was given dry food but no water. After two groups had been under this regime from one to thirteen days, they were killed and the posterior lobes removed. Five lobes of each group were extracted in saline for bioassay; the other two were prepared for microscopic study according to Gersh. The antidiuretic potency of the normal and dehydrated rats' posterior lobes was estimated from the concentrating effect on the urine of dogs with diabetes insipidus. It was found that the antidiuretic potency of the posterior lobe decreased rapidly with dehydration of the rat, so that by the thirteenth day without water the posterior lobe contained less than three per cent of its normal antidiuretic hormone.

There was, however, no correlation between the hormonal and osmiophilic droplet content of the posterior lobes.

Pituitaries from rats having a permanent polyuria after pituitary stalk section were cut in a sagittal plane. One half was assayed, the other studied according to the technique of Gersh. The antidiuretic assay indicated a potency of much less than one per cent of normal, but the stained portion of the gland contained an enormous number of osmiophilic droplets.

The renal excretion of chloride by the dog. RUTH STILLMAN HARE (by invitation) and KENDRICK HARE. *Department of Anatomy, Cornell University Medical College, New York City.*

The excretion of chloride was followed in the unanesthetized dog before, during, and after the infusion of 0.9 per cent to 10 per cent NaCl solutions. Glomerular filtration was measured by creatinine clearances. After the

chloride infusion was begun, glomerular filtration rose to values which in some cases were twice normal. Maximal tubular reabsorption of glucose did not increase with increasing glomerular filtration. This observation indicates that no additional nephrons were recruited.

On the assumption that the concentration of chloride in glomerular filtrate is the same as that in plasma, and that the creatinine clearance is a measure of glomerular filtration, the amount of chloride filtered per minute was calculated. The amount excreted per minute was determined by urine analysis. The difference between these two quantities is the amount reabsorbed.

The infusion of NaCl solution increased the amount of chloride filtered by increasing both the glomerular filtration and the plasma chloride. When the filtration rate of chloride was increased above the pre-infusion level, approximately 35 per cent of the increase in chloride filtered was excreted.

Apparently, chloride U/P ratios were not the determining factor in tubular reabsorption of chloride, since they continued to increase throughout the experiment.

Since only sodium chloride was administered, this data might apply to the mechanism for the excretion of chloride, or sodium, or sodium chloride.

Chloride excretion in the diabetes insipidus dog is like that in the normal dog and was not increased by 0.1 to 10 milliunits of pitressin, although marked antidiuresis was obtained.

The fate of fluids injected in animals suffering from traumatic shock. HENRY N. HARKINS, R. T. BOALS and C. FRANK CHUNN (introduced by L. R. Dragstedt). *Department of Surgery, Henry Ford Hospital, Detroit, Mich.*

It is well known that when shock has advanced to a certain point it becomes irreversible and remedial measures, no matter how thorough, are of no avail. This is true even in cases of shock from simple hemorrhage where, if the blood pressure has been allowed to remain low for a prolonged period, replacement of even more blood than was lost does not cure.

In animals in which shock has been produced by localized trauma, death usually follows the loss of about three per cent body weight of plasma-like fluid into the injured area (as determined by bisection experiments, or in the case of peritoneal trauma, by paracentesis). In other animals given massive doses of normal salt solution intravenously, this local loss was nine to ten per cent body weight in many cases. This indicates a marked local loss of fluid given therapeutically. Other animals given plasma or concentrated plasma showed a marked increase in the extent of local loss over the untreated series but not nearly as much as following the use of saline. In the cases given saline the protein content of the edema fluid indicated a greater absolute loss of plasma protein than in the untreated animals. In all experiments only parts of the fluid administered could be accounted for as a local loss at the site of trauma, indicating a generalized increased capillary permeability similar to that postulated by Moon (Ann. Int. Med. **13:** 451, 1939).

Animals with marked hemoconcentration and shock were given concentrated plasma with sudden dilution following injection of the plasma, but immediate reconcentration in severe cases. Normal animals showed

an initial concentration of the blood following concentrated plasma administration with a subsequent return to the normal level. This difference in response is of interest and would indicate a difference in capillary permeability in the shocked animals as compared to the normal.

Conclusions: 1. Quantitative evidence is offered to show that following fluid administration in shock, local fluid loss is increased, especially when crystalloids are given. 2. Some fluid is lost from the blood stream throughout the body. 3. Concentrated plasma does not produce a progressive blood dilution when severe shock is present.

Polarization effects in mammalian hearts related to the establishment of idioventricular rhythms and fibrillation.[1] A. SIDNEY HARRIS and GORDON K. MOE (by invitation). *Department of Physiology, Western Reserve University Medical School, Cleveland, O.*

Effects of cathodal and anodal polarizing currents (indifferent electrode in body wall) on ventricular activity were studied by recording simultaneously from three loci, using three string galvanometers. The leads were riding electrodes consisting of proximate bipolar Ag·AgCl tips mounted in a small lucite block and held by a flexible resilient suspension. This type of electrode leads significantly from a small localized area only. When high stimulating voltages were applied through electrodes 1 cm. apart, 1 cm. from the leads, potential differences recorded by the leads were about 0.05 per cent of those applied.

Polarizing currents insufficient to induce beats of ventricular origin cause undulations to appear in the electrograms. Isoelectricity may disappear entirely. The undulations are not regular. In cathodal polarization they occur roughly at multiples of 40 msec. Anodal polarization presents a different pattern but undulations occur and the tempo of prominences is similar. Strong currents produce tachycardia characterized by longer deflections and irregularities. During anodal polarization one of the prominent features comes at a time corresponding with the T wave, and when there are coupled beats, the second occurs during the T wave of the first. During polarization tachycardia the intervals between beats, though varying widely especially in anodal records, appear related to those of the undulation. During cathodal tachycardia the rate may be accelerated by 40 per cent, but remains relatively constant. The sequence of R deflections at three points on the ventricle shift frequently with relation to each other, and may bear no relation to the normal sequence. Anodal tachycardia has a very irregular rhythm usually characterized by accelerating runs of beats each run terminated by a pause, or in some cases by fibrillation.

Fibrillation may be induced by anodal or cathodal polarization, with the anodal possibly the more efficacious. Fibrillation induced by anodal or cathodal polarization or by brief stimuli usually begins by rapidly accelerating tachycardia which fails to be blocked. During these runs there is relative constancy in the order of excitation at a series of points.

The action of Integerrimine, Jacobine, Longilobine, Ridelliine, Senecionine and Spartioidine. PAUL N. HARRIS (by invitation), ROBERT C. ANDERSON (by invitation) and K. K. CHEN. *Lilly Research Laboratories, Indianapolis, Ind.*

[1] Aided by a grant from the John and Mary R. Markle Foundation.

The above alkaloids were isolated by Manske from *Senecio* species (Canad. J. Research **5:** 651, 1931; *Ibid.* B, **14:** 6, 1936; *Ibid.* B, **17:** 1, 1939). Experiments were made in 373 mice. The median lethal doses by intravenous injection were found to be as follows:

ALKALOID	MEDIAN LETHAL DOSES ± STANDARD ERROR
	mgm. per kgm.
Integerrimine	78.32 ± 3.05
Jacobine	77.11 ± 2.86
Longilobine	77.85 ± 3.33
Ridelliine	108.8 ± 3.06
Senecionine	64.12 ± 2.24
Spartioidine	80.39 ± 1.93

Like retrorsine and seneciphylline, these alkaloids caused delayed deaths in many animals, and produced various degrees of hepatic necrosis and congestion. Ascites or hydrothorax, or both, occurred in slightly less than half of the mice. The sinusoidal congestion of the liver was diffuse in most cases, but occasionally it became so extreme in places that it resembled cavernous hemangioma. Similar pathological changes were observed when large fractions of the lethal dose of jacobine were repeatedly administered per os or by subcutaneous injection over a period of days. There were some differences in the action of the six alkaloids, but they were chiefly quantitative in nature. In no instance was cirrhosis of the liver noted similar to that reported for the cattle disease prevalent in South Africa.

The differentiation of urogastrone and pituitrin.[1] STANLEY C. HARRIS (by invitation), JOHN S. GRAY and E. WIECZOROWSKI (by invitation). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Because both pituitrin and urogastrone are found in urine and both are capable of inhibiting gastric secretion and motility, their physiological effects have been compared in order to investigate the possibility of their being identical.

Approximately equal degrees of inhibition of the gastric secretory response of total pouch dogs to histamine was obtained with 3 mgm. of urogastrone and 4 units of pituitrin. Equal inhibition of gastric motility, stimulated and maintained in trained unanesthetized dogs by distension with the recording balloon, was obtained with 3 mgm. of urogastrone and 0.1 unit of pituitrin. Diuresis induced in a trained dog with a bladder fistula by the oral administration of 250 cc. of water was not altered by 3 mgm. of urogastrone, whereas 0.001 unit of pituitrin produced a striking inhibition. Urogastrone in 6 mgm. doses is without effect on blood pressure, whereas 4 units of pituitrin produced an easily demonstrable rise in blood pressure in anesthetized dogs.

The lack of parallelism in the physiological properties of pituitrin and urogastrone indicates that they are separate entities.

[1] Aided in part by a grant from the Committee on Endocrinology of the National Research Council.

The central pathway for the jaw jerk. FRANK HARRISON and KENDALL B. CORBIN (introduced by Lathan A. Crandall, Jr.). *Division of Anatomy, University of Tennessee, Memphis.*

Recent work from this laboratory indicating that the mesencephalic root and nucleus of the trigeminal nerve mediate proprioceptive impulses from the muscles of mastication does not support the findings of Bremer (1923) and Rioch and Lambert (1934). These latter workers concluded that the mesencephalic root was not a part of the jaw jerk reflex arc.

With the Horsley-Clarke instrument and cathode ray oscillograph the mesencephalic root of the fifth cranial nerve in the cat was located, and then stimulated with faradic or destroyed with direct current. From a few minutes to as much as 23 days later, each animal was decerebrated at the thalamic level and the jaw reflexes studied. The brain of each animal was examined microscopically and the site of the lesion correlated with changes in reflex activity. In none of the animals was there damage to the motor nucleus, chief sensory nucleus or spinal nucleus of the fifth nerve. Lesions were placed only on one side and the reflex activity of the normal side was used as a control. Presence or absence of muscular contraction was determined by recording action potentials with needle electrodes inserted into the masticator muscles.

The jaw opening reflexes to faradic stimulation of gums and palate were not depressed by lesions in the mesencephalic root. Jaw closure to tapping the teeth (which sometimes elicits jaw opening) was abolished by lesions in the caudal part of the mesencephalic root. The jaw jerk, a stretch reflex, was abolished by complete destruction of the homolateral mesencephalic root or was diminished if there was only partial damage. If only the most caudal part of the mesencephalic root and nucleus remained intact, the jaw jerk might still be elicited although attenuated. Destruction of the root did not abolish the jaw closing that accompanies reflex swallowing.

The role of collateral fibers from the mesencephalic root to the motor nucleus and the results of stimulation of the trigeminal nuclei will be presented. It is concluded that the afferent pathway for the jaw jerk is the mesencephalic root of the fifth nerve.

Water metabolism in the chicken with special reference to absorption from the cloaca. WILLIAM MILTON HART (introduced by Hiram E. Essex). *Division of Experimental Medicine, The Mayo Foundation, Rochester, Minn.*

The ureteral orifices of a series of adult chicken hens were exteriorized by operative means so that the urine was voided directly to the outside without having had contact with the cloacal or gut mucosa. In another series of birds an artificial anus was established by the method of Milroy in order to obtain urine having had contact with the cloacal mucosa. Some of the birds in which the orifices of the ureters were exteriorized have been in the laboratory for eighteen months and are still in excellent physical condition.

Studies of water consumption on the same birds before and after surgical procedures showed that birds with exteriorized ureteral orifices usually drink more water following the operation. After a week's time on a diet without added salt, the blood chloride fell from a normal 400 mg. per cent to about 360 mgm. per cent and the hematocrit rose from a normal 30 to 40 per cent to as high as 57 per cent. When 1 per cent salt was added to

the diet the blood chloride and the hematocrit returned to normal. The bird also regained the weight lost from dehydration. When drinking water is withheld, normal birds lose an average of 30 grams of body weight per day. Birds with exteriorized ureteral orifices lose weight twice as rapidly as normal birds.

The hens with artificial anuses do not differ greatly from birds with exteriorized ureters in regard to water consumption. However, hens with artificial anuses show no reduction of blood chlorides which would indicate that they probably absorb chloride from the rectum.

It is concluded that water and salt are absorbed from the rectum of the fowl and that absorption from the proctodaeum is too small to be of consequence.

Dark adaptation of single visual sense cells. H. K. HARTLINE and R. MCDONALD (by invitation). *Cornell University Medical College, New York City, and the University of Pennsylvania, Philadelphia.*

Changes in sensitivity of visual sense cells in the eye of *Limulus* during dark adaptation have been followed by recording the impulses discharged in single optic nerve fibers in response to test flashes of light of fixed intensity and duration. Immediately following exposure to light the number of impulses discharged in response to the test flash is less than the number elicited by this same flash when the sense cell is completely dark-adapted. The higher the intensity and the longer the duration of the preceding adapting exposure, the greater is this initial reduction in sensitivity of the receptor. Recovery of sensitivity during dark-adaptation is rapid initially and approaches the dark adapted value asymptotically. Measured at a fixed level of sensitivity below the dark-adapted value, the rate of recovery following different amounts of light adaptation is slower the greater the initial light adaptation. Recovery following prolonged illumination at low intensity is slower than recovery following a short, bright flash, even though the initial reduction in sensitivity is the same in both cases. These results can be explained qualitatively by a visual cycle of photolysis and regeneration such as that proposed by Wald.

Not only is the total number of impulses in response to a given test flash decreased following light adaptation, but the frequency of the discharge is also reduced. However, the frequency is much less affected than is the total number of impulses; the latency of the response is only slightly affected. Thus it is not possible to match responses obtained under different conditions of adaptation by merely altering the intensity of the test flash until the number of impulses is equal. During light- and dark-adaptation, therefore, other changes must take place in the visual sense cell than simple alterations in sensitivity due to altered concentration of photosensitive substances.

Cortical responses to click stimulation. J. E. HAWKINS, JR. (introduced by H. Davis). *Department of Physiology, Harvard Medical School, Boston, Mass.*

Responses of the auditory cortex of the anesthetized cat (Evipal, pentobarbital sodium, ether) to click stimuli have been analyzed by several methods.

Cortical responses (monopolar recording) are preceded by small waves

spreading from lower nuclei of the auditory pathway and a small positive wave from the auditory radiations. Two primary response patterns of 6–10 msec. latency are distinguished. Pattern I, occuring regularly in the anterior ectosylvian gyrus, consists of a negative wave of 100–200 μV. lasting 15–20 msec., followed by a smaller slower positive wave (50–60 msec.). Pattern II, predominant in the middle ectosylvian gyrus, consists of a positive wave of 200–500 μV. lasting 20–25 msec., followed by a negative wave of almost equal amplitude and 50–80 msec. duration; this is usually succeeded by a slow positive wave of small amplitude. Combinations of the two patterns may occur.

Penetration of the cortex by 1 mm. or less causes response pattern I to replace pattern II, and after thermocoagulation of the outermost layers of the cortex only pattern I can be recorded from the surface. Therefore elements essential for pattern II are located in the outer layers, while those responsible for pattern I are more widely distributed

The large negative wave of pattern II is much reduced if stimuli are more rapid than 10 per sec. It is eliminated by increasing the depth of anesthesia with ether or by local application of pentobarbital to the cortex, and is enhanced by strychnine. It probably represents an efferent discharge from the cortex.

Under ether a second positive wave (peak time 35–40 msec.) follows the first positive component of pattern II (peak time 16–17 msec.). During a change to Evipal anesthesia the second positive wave falls at progressively longer intervals after the first, becoming variable in appearance and latency (100–200 msec.). It has several points of similarity to the secondary response described by Forbes and Morison.

A click applied to the ear simultaneously with an electrical stimulus to the cortex evokes in the opposite cortex a response which is found to be the simple summation of the responses to the two stimuli applied separately.

Methods useful to the physiological study of the biodynamics of stance. F. A. Hellebrandt, L. E. A. Kelso (by invitation), Rozell Henkel (by invitation) and Corrine Fries (by invitation). *University of Wisconsin, Madison.* (Demonstration.)

Oscillograms of the shifts in the center of gravity which occur in the vertical orientation planes during standing indicate a high degree of constancy in the natural stance patterns of man. When projected into the base of support they are slightly eccentric, usually being forwardly displaced and asymmetric. The asymmetry is predominantly to the left and may be related to handedness, footedness and strength which are being measured. The repeatability of oscillographic patterns made periodically during one hour of uninterrupted standing suggests that within these limits, standing is indefatigable. By synchronizing the center of gravity observations with simultaneous biplane photography a record may be obtained of the influence of varying gravitational stresses upon the postural disposition of the multijointed skeletal parts. If the postural sway is excessive in the transverse vertical plane, shifting physiological curvatures of the vertebral column, compensatory in nature, may be observed. By superimposing successive photographs taken at 5, 10 or 15 second intervals during relaxed and comfortable standing, the major compensatory adjustments may be localized. These are most conspicuous at the knee and in

the region of the lumbar spine, suggesting that the body does not sway *in toto* over the ankle joints.

Chemical temperature regulation of the dog. ALLAN HEMINGWAY. *The Medical School, University of Minnesota, Minneapolis.*

Chemical temperature regulation, defined as the control of heat production of an animal in response to cold, has been divided into two parts, namely, 1, a preshivering component, and 2, shivering. Experiments have been performed to determine whether or not the preshivering component exists in normal dogs and its relation to shivering.

Dogs trained at least two months were used. In an experiment each animal was placed in a metal box with hollow walls through which water of controlled temperature circulated to produce a measurable and uniform rate of cooling. The dog's head was sealed in a head mask connected to a metabolism apparatus for measuring rate of oxygen consumption, CO_2 and water production. After a one hour rest the metabolism was measured, I, during a basal period, II, during a cooling period in which the metal box temperature was lowered to produce shivering in 20 to 40 minutes, and III, during the subsequent period of shivering. Shivering was determined by electrical recording of amplified muscle action currents, a sensitive mechanical recorder and visual inspection. Skin and rectal temperatures, respiration and heart rates were also measured. In control experiments basal conditions of temperature were maintained throughout the three periods.

Twelve to sixteen experiments were performed on each of two well trained and cooperative dogs. Of special interest were the metabolism and electrical changes during the second (preshivering) period. During this period one dog exhibited an average increase of six per cent in metabolism while the other dog increased his metabolism an average of eleven per cent. These differences, which are significant in regard to number of experiments and controls, lead to the conclusion 1, that a preshivering rise in metabolism occurs; 2, that the preshivering metabolism is small and varies with different animals, and 3, it is not directly related to the shivering process.

The infra-red absorption spectra of estrogens, androgens and related steroids.[1,2] CARL M. HERGET (by invitation) and EPHRAIM SHORR. *The Russell Sage Institute of Pathology in affiliation with The New York Hospital and Department of Medicine, Cornell University Medical College, New York City.*

The infra-red spectra of a number of androgens, estrogens and related steroids in the solid state have been obtained using the J. D. Hardy infra-red spectrophotometer. Unlike visible and ultraviolet absorption spectra, which reflect the electronic state of the molecule, the appearance of the infra-red absorption is determined by atomic groupings. This region, therefore, could be expected to yield unique absorption spectra. Other advantages of the method are its rapidity, only one scanning being necessary for a complete analysis and the small samples required (3–5 mgm.) which are unaffected by the analysis.

Among the compounds studied in the pure state were androsterone,

[1] Aided by the Wyeth Endocrine Fund.
[2] The compounds studied were generously supplied by Dr. Erwin Schwenk of the Schering Corporation.

dehydroandrosterone, androstenedione, isoandrosterone, dehydroisoandrosterone, testosterone, androsterone benzoate, alphaestradiol dipropionate, cholestenone, desoxycorticosterone, pregnenolone, progesterone, cholestanone and cholesterol.

Results: All the above compounds have, in the solid state, unique infrared absorption spectra, except spacial isomers which have similar spectra. Experiments are in progress which promise to permit the differentiation of spacial isomers by carrying out the observations in appropriate solvents. From these spectra it can be determined with certainty whether certain groups, such as the ketonic and hydroxyl, are present. Ketonic and hydroxyl groups on the 3-carbon atom can usually be distinguished from those on the 17-carbon atom.

This method is applicable to the analysis of compounds present in biological fluids.

The effects of renin and of angiotonin on the renal blood flow and blood pressure of the dog. J. F. HERRICK, A. C. CORCORAN and H. E. ESSEX. *Division of Experimental Medicine, The Mayo Foundation, Rochester, Minn., and The Lilly Laboratory for Clinical Research, Indianapolis City Hospital, Indianapolis, Ind.*

Renin and angiotonin have been observed to increase arterial pressure and decrease renal blood flow in studies in which blood flow was measured indirectly, either by clearance and extraction of phenol red and inulin through the explanted dog's kidney, or by the clearance of diodrast (Corcoran and Page). Both substances were given intravenously by slow infusion in these experiments. Steele and Schroeder have studied the effects of single intravenous injections of renin on the blood flow in the renal artery and other arterial vessels of the dog by the means of thermostromuhr and have reported that such injections were usually followed by increased renal blood flow.

These discordant results prompted the present investigation, in which the direct current thermostromuhr was used as the measure of arterial flow. A series of experiments were performed on seven dogs, four of which were anesthetized with pentobarbital sodium during the observations of renal blood flow and femoral arterial pressure. Femoral blood flow was observed in two of these dogs. The remaining three dogs had been trained and the thermostromuhr unit applied several days before observations of blood flow and blood pressure.

Single injections or slow intravenous infusions of renin or angiotonin produced a marked increase in blood pressure and a decrease in renal blood flow. The response to renin was usually delayed and prolonged, whereas the effect of angiotonin was more immediate and relatively transient. The magnitude of the response depended on the dose administered. Varying degrees of refractoriness, manifested in total or partial lack of response to subsequent injections, appeared in all but one instance after slow infusions of a few cc. of either substance. One trained dog failed to develop the refractory state and continued to show marked reductions of renal blood flow and increases of arterial pressure with succeeding injections of angiotonin.

The effect of renin and angiotonin on the blood flow in the femoral artery was initially diphasic, although a significant increase of blood flow occurred later.

Comparative effects of stimulants on infant and adult cerebral tissues.[1] HERMAN HERRLICH (by invitation), JOSEPH F. FAZEKAS (by invitation) and HAROLD E. HIMWICH. *Department of Physiology and Pharmacology, Albany Medical College, Union University, Albany, N. Y.* (Read by title.)

The cerebral metabolism of infant and adult rats was compared in the presence of various stimulants: methylene blue, p-phenylenediamine, potassium, and increase of temperature. Minced tissues were suspended in Ringer's solution buffered with phosphate at pH 7.4 and placed in a Warburg respirometer.

Dixon has previously demonstrated that the oxygen consumption of adult cerebral tissues increases about 50 per cent when 0.1M K is added to Ringer's solution. The present results reveal that the response to K is slight in rats 1 to 10 days of age. The stimulatory effect of K increases with age and in rats 25 days old is of the same magnitude as the adult. In a similar fashion the increase of oxygen consumption with a rise of temperature is greater in the adult than in the infant. With a rise of temperature from 38° to 42° the acceleration of metabolism per hour is 17 per cent for the adult and 10 per cent for the infant. The same amounts of p-phenylenediamine are oxidized more rapidly by the adult than by the infant brain. The acceleration of metabolism in the adult decreases after 20 minutes while in the infant the increase, though smaller, is maintained throughout the observation period of one hour. With glucose as substrate the time required for the decolorization of 2 cc. of 0.0002 per cent methylene blue by 500 mgm. of cerebral tissue of adult rats is 14.8 minutes (average of 10 observations) and 34.1 minutes for the infant (average of 9 observations). The results with methylene blue and p-phenylenediamine indicate lower concentrations of dehydrogenase and oxidase in the young. The lesser stimulatory effects of K and increase of temperature suggest that in the young these enzymes may be working closer to their maximum activity.

Vascular reactions of the finger to cold.[2] A. B. HERTZMAN, L. W. ROTH (by invitation) and J. B. DILLON (by invitation). *Department of Physiology, St. Louis University School of Medicine, St. Louis, Mo.*

The vascular reactions of the finger to its local chilling, previously described by Lewis and others, have been examined by means of the photoelectric plethysmograph.

On adequately cooling a single finger, vasoconstriction occurs not only there, but also in the "control" fingers of both hands. The constriction occurs at once and independently of the presence or absence of pain. Usually, it is more profound and lasts much longer in the chilled finger, but during its existence vasoconstrictor activity is enhanced in the "control" fingers of both hands even though the chilled finger becomes numb from cold. After a variable period (2 to 8 min. or more), dilatation begins in the chilled finger, reaching its maximum in a progressive manner. During the early part of the dilatation and also during the latter part of the initial constriction, vasoconstrictor paralysis (complete or partial) is often but not

[1] Aided by a grant from the Child Neurology Research (Friedsam Foundation).
[2] Aided by a grant from the Committee on Therapeutic Research, Council on Pharmacy and Chemistry, American Medical Association.

always present in the chilled finger. Often this shows in the form of a progressive or delayed constriction contrasting sharply with that in the "control" finger. The period of dilatation ends (when it does) only through the action of vasoconstrictor discharge.

These and other similar observations emphasize the importance of vasomotor reflexes elicited by the thermo-sensory and other stimuli, in these reactions to cold. They neither affirm nor deny a possible participation of arterio-venous anastomosis in the reactions. It appears, however, that the pad arteries dominate the plethysmograms.

In the normal subject, the form of the pulse wave in the chilled finger is not appreciably affected either during the constriction to cooling or in the subsequent reaction of dilatation. The propagation of the wave may or may not be slower in the chilled finger than in the "control."

In a few subjects, otherwise normal, chilling of the finger resulted in a normally appearing finger pulse becoming similar to that observed in cases of hypertension and arteriosclerosis (described elsewhere). This change was most noticeable during the dilatation. The reasons for this shift have not been determined but are apparently related to the elevation of predicrotic wave—"Zwischenschlag"—so that it becomes the crest of the wave.

The relation of various hypothalamic lesions to adiposity and other phenomena in the rat. ALBERT HETHERINGTON (introduced by S. W. Ranson). *Institute of Neurology, Northwestern University Medical School, Chicago, Ill.*

Study of a second series of 22 rats in which lesions of various sizes have been placed in several different parts of the hypothalamus with the Horsley-Clarke instrument leads to the following tentative conclusions.

Obesity is not produced by: 1, bilateral interruption of the fornix, with small damage to surrounding tissue in the anterior hypothalamus; 2, medium-sized symmetrical lesions of the caudal half of the anterior hypothalamic area and rostral ends of the ventromedial hypothalamic nuclei; 3, large lesions destroying most of the mammillary body; 4, large lesions destroying almost all of one side of the hypothalamus.

Obesity can be produced by: 1, fairly symmetrical lesions limited to the ventromedial hypothalamic nuclei, the medial halves of the lateral hypothalamic areas, and the ventral portions of the dorsomedial hypothalamic nuclei; 2, fairly symmetrical lesions limited to the caudal ends of the ventromedial and dorsomedial hypothalamic nuclei, the premammillary group of nuclei, and the adjacent parts of the lateral hypothalamic area. A considerable degree of symmetry in the location of the lesions on the two sides appears to be important for producing obesity. The possibility remains that destruction of other regions of the hypothalamus as yet unexplored may cause the disturbance.

Completely normal sex cycles seldom occurred in either obese or non-obese operated animals; commonest were cycles irregular in length, and prolongation of some phase (usually atypical) of the cycle other than that of dioestrus. Diabetes insipidus was found only in animals having lesions immediately caudal and medial to the supraoptic nuclei, or in the median eminence. Food intake and activity cage experiments revealed that an enormous decrease of activity on the part of the operated animals was probably more responsible than augmented food intake for the obesity.

However, spontaneous running was greatly reduced even in non-obese operated animals.

In most of the cases in which serial sections of the hypophyses of the fat animals were examined the lesion did not enter the pituitary. Histological differences between these glands and those of normals are not conspicuous.

The control of glomerular function in the seal (Phoca vitulina, L.). EDWIN P. HIATT and STANLEY E. BRADLEY (introduced by H. W. Smith). *Department of Physiology, New York University College of Medicine, New York City, and the Mount Desert Island Biological Laboratory, Salsbury Cove, Maine.*

Marine mammals apparently meet their water requirements entirely from the water derived from their food. It is to be suspected that such animals will show specialization of renal function to conserve their meager supply of water. We have endeavored to determine the nature of this specialization by measuring the glomerular filtration rate and the renal plasma flow in young harbor seals.

We have taken the creatinine clearance as a measure of the glomerular filtration rate, and have considered the diodrast clearance to be a very close approximation of the renal plasma flow. For each experiment an animal was removed from the floating trap in which they were kept and strapped on its back in a trough. Creatinine was administered by stomach tube about an hour before collections were begun. Diodrast was injected under the loose skin of the flippers about 40 minutes later. Urine was collected by catheter and blood was taken in an oxalated syringe from the flipper veins.

Shortly after ingesting a kilogram of herring, the filtration rate and the renal plasma flow of these animals increased markedly, in some instances as much as four times the fasting level. This increase lasted for several hours after feeding but disappeared within twenty-four hours.

The creatinine/diodrast clearance ratio remained almost constant at about 0.30 over the whole range included in our experiments. This indicates that the increased blood flow is due, in part, to a dilation of the afferent renal arterioles. This ratio is constant only if care is taken to avoid trauma and excitement. When such precautions are not observed the ratio is higher, indicating a constriction of the efferent renal arterioles.

Tolerance of the newborn to hypoxia and anoxia.[1] HAROLD E. HIMWICH, F. A. D. ALEXANDER (by invitation) and JOSEPH F. FAZEKAS (by invitation). *Department of Physiology and Pharmacology, Albany Medical College, Union University, Albany, N. Y.*

This report presents evidence that the infant is more resistant than is the adult both to hypoxia and hypoglycemia. Mature rats placed in a jar containing nitrogen stop respiring in from 3 to 4 minutes and cannot be resuscitated while newborns in the same jar live 50 minutes. These results were observed at room temperature, 24°C. When the environmental temperature is raised to 35°C., the newborn survives 20 minutes. In an atmosphere of nitrous oxide the results are similar but in carbon dioxide respiratory efforts persist for 25 minutes only and in cyclopropane for 15

[1] Aided by a grant from the Child Neurology Research (Friedsam Foundation).

minutes. Sensitivity to these gases increases with age and at approximately 12 days of age the survival period is the same as that of the adult.

Experiments on puppies yield comparable results. Newborn dogs survive from 24–35 minutes and samples of arterial blood drawn at different intervals starting as soon as 5 minutes after the initiation of the inhalation of nitrogen reveal no oxygen within the error of the method. In those experiments in which the anoxia was prolonged until the animal succumbed, electrocardiographic studies disclosed that the heart continued beating after respiration had ceased. The heart of the puppy, like its central nervous system, is more resistant to anoxia than the adult. Further experiments on puppies disclose that these animals can tolerate a mixture of 5 per cent oxygen in nitrous oxide for much longer periods than can adult animals. Puppies 1 to 12 days of age withstand this mixture for at least 3 hours despite blood oxygen levels persisting between 4 and 5 volumes per cent. In the same gas mixture adult animals survive only 10 to 15 minutes. Newborn rats may live in a mixture of about 5 per cent oxygen and 95 per cent nitrous oxide for more than 7 hours. Adult rats tolerate the same mixture for less than 20 minutes.

The low cerebral metabolic rate of the infant can explain the prolonged survival time only in part since the metabolic rate remains low while tolerance to anoxia rapidly decreases. Factors which may contribute to make the newborn relatively tolerant to anoxia are 1, low cerebral metabolic rate; 2, poikilothermia, and 3, anaerobic source of energy.

Hypoglycemia in the infant rat.[1] HAROLD E HIMWICH, JOSEPH F. FAZEKAS (by invitation) and F. A. D. ALEXANDER (by invitation). *Departments of Physiology and Pharmacology and Anesthesia, Albany Medical College, Union University, Albany, N. Y.* (Read by title.)

The present experiments demonstrate that infant rats tolerate hypoglycemia better than adults. Adult and infant rats each received the same dose of insulin, 10 units. The adult rats became comatose and sometimes died while the young resisted coma and survived from 5 to 10 hours despite the fact that on a weight basis their dose was at least 10 times greater. Determinations of blood sugar reveal that the prolonged survival period of the infant rat is not due to insulin resistance for their blood sugar assumed hypoglycemic levels. Anoxia superimposed in hypoglycemia acts synergistically in the newborn as in the adult and the animal succumbs sooner than with either alone. The survival time of newborn rats injected with insulin and subjected to the inhalation of nitrogen was reduced from 50 minutes to 25 minutes. Even when insulin is not administered glucose injected intraperitoneally or subcutaneously protects rats exposed to anoxia. The survival period of infant animals 8 to 10 days of age is prolonged from 7 to 15 minutes, if they receive glucose before the inhalation of nitrogen.

Some effects of CO_2, anoxia and alcohol on respiration. FRED A. HITCHCOCK. *Department of Physiology, The Ohio State University, Columbus.*

Experiments have been carried out in which the effects of high concentrations of CO_2, low concentration of O_2, and the ingestion of alcohol on the respiration of human subjects have been compared. The effect of super-

[1] Aided by a grant from the Child Neurology Research (Friedsam Foundation).

imposing alcohol on CO_2 increase and anoxia has also been noted. When the CO_2 of the inspired air is increased to about 7.5 per cent the ventilation volume was raised from 200 to 450 per cent. Usually both tidal air and rate were increased. Lowering the O_2 in the inspired air to about 6 per cent increased the ventilation volume from 90 to 125 per cent. This effect was exclusively on the volume of the tidal air. The ingestion of moderate amounts of alcohol (blood concentration ranging from 0.5 to 0.8 mgm. per cc.) acted first as a stimulant to respiration and then as a depressant. The period of stimulation lasted for not more than 30 minutes. Following this initial period of stimulation alcohol decreased the stimulating effect of both CO_2 and anoxia. The decrease was most pronounced in the case of anoxia which seems to indicate that the depressing effect of alcohol is more pronounced on peripheral reflexes than on the respiratory center itself.

Revival of mammalian sperm after immersion in liquid nitrogen. HUDSON HOAGLAND and GREGORY PINCUS. *Clark University, Worcester, Mass.*

Luyet and Hodapp (Proc. Soc. Exper. Biol. and Med. **39:** 433, 1938) reported that an appreciable percentage of frog sperm, after partial plasmolysis, can survive immersion in liquid air. Rapid cooling and warming is necessary, the sperm passing in and out of the vitrified state without having time to freeze (i.e., crystallize). These workers could not demonstrate revival of similarly treated rat sperm. Shettles (Am. J. Physiol. **128:** 408, 1940) obtained revival of a few per cent of human seminal sperm after immersion of small samples in capillary tubes in liquified gases.

We have used human seminal sperm from the same donor in a series of experiments attempting to improve the viable yield after immersion in liquid nitrogen followed by rapid warming. Films of seminal sperm several hours after emission, trapped on small wire loops and dipped in liquid nitrogen, showed, in general, less than one per cent of motile sperm revivable. This yield could be improved to several per cent with less than hour-old suspensions. Our best results to date have been obtained by mixing the fresh human seminal sperm samples with rabbit serum and entrapping them in thin bubble films blown from a fine pipette. These bubbles in the form of an air emulsion, transferred by wires to liquid nitrogen, have produced yields as high as 40 to 50 per cent of viable sperm after rapid warming. These revived sperm show the same type of motility as do controls.

Sperm from the vas deferens of 14 rabbits have been studied to date under a variety of conditions of pretreatment. We have obtained only about 0.1 per cent of live rabbit sperm after bubble film exposure to liquid nitrogen from two animals. On one occasion the pretreatment consisted of two minutes' plasmolysis of rabbit serum sperm suspended in Ringer solution 5 times the isotonic concentration. A similar yield was obtained by plasmolysing a suspension without serum for 10 minutes in a Ringer solution of twice the normal Ca content.

Correlation between the secretory power of the frog kidney and the molecular configuration of organic compounds. RUDOLF HÖBER. *Department of Physiology, School of Medicine, University of Pennsylvania, Philadelphia.*

In previous experiments on the isolated Ringer perfused frog kidney it has been found that the sulphonic acid azodyestuffs undergo secretory transfer across the proximal tubules, provided the sulphonate groups are attached to one of the molecular halves of these dyes, whereas the distribution of the sulphonate groups to both halves, in general, impedes the secretion. It was concluded that a polar-nonpolar or a hydrophilic-organophilic structure is a prerequisite for the attachment of the dye molecule to the surface of the kidney cells.—These previous studies have been extended with the use of more simple organic compounds. The kidney was supplied with only one half of the azodye molecules and this half, upon reappearing in the secretion, was coupled in a diazo reaction to the other half and determined colorimetrically. The following results were obtained: the monosulphonates of naphthylamine, naphthol and aminonaphthol, irrespective of the mutual location of the NH_2, OH and HSO_3 groups, appear in the secretion at a higher concentration level than that in the perfusion fluid, whereas the disulphonates fail to become accumulated. It is suggestive to interpret these results in a way similar to that proposed with regard to the behavior of the azodyestuffs: organotropic affinities are exhibited by the naphthalene ring system as well as by the NH_2 and OH groups, hydrotropic affinities by the sulphonates. If the opposing forces balance each other, there is an attachment of the molecule at the interfacial boundary, a preliminary step in the active transfer. This hypothesis is supported, for instance, by comparing 1, aminonaphthalenesulphonic acid; 2, sulphanilic acid; 3, sulphanilyl-sulphanilic acid; 4, sulphanilyl-sulphanilamide; 1 and 3 were concentrated in the secretion, 3 and 4 were not.

A further study of pupillary responses to electrical stimulation of the fore- and mid-brain. ROBERT HODES (by invitation) and H. W. MAGOUN. *Institute of Neurology, Northwestern University Medical School, Chicago, Ill.*

Since recent work has emphasized the importance of oculomotor inhibition in reflex dilatation of the pupil, it is desirable to determine the central pathways concerned. As a preliminary step, the regions of the upper brain-stem of the cat which upon stimulation yield pupillary dilatation have been explored with the Horsley-Clarke technique, and the oculomotor-inhibitory and sympathetic-excitatory areas compared.

Pupillary dilatation effected by oculomotor inhibition is elicited from a widespread area including the medial part of the basal forebrain, the entire extent of the hypothalamus with a dorsal projection into the midline region of the thalamus, and in the midbrain from the central gray and the tegmentum.

Dilatation of the pupil effected by sympathetic excitation was not obtained from stimulating the basal forebrain in front of the optic chiasma, but was elicited from the antero-posterior extent of the hypothalamus. In the midbrain, responsive points were confined to the tegmentum.

Concomitant observations of other sympathetic effects (piloerection, retraction of the nictitating membrane, rise of blood pressure) reveal a distribution of excitable regions analogous to those for sympathetic dilatation of the pupil, rather than to those for dilatation by oculomotor inhibition.

Although these results do not define the central pathways involved in

reflex, emotional and other pupillary dilatation, they provide a foundation for further study of such mechanisms.

Cholinesterase in the spinal cord of cats after section of dorsal roots.[1] E. C. HOFF and D. NACHMANSOHN. *Laboratory of Physiology, Yale University School of Medicine, New Haven, Conn.*

If the activity of cholinesterase is intrinsically related to nervous function, degeneration of nerve fibers should lead to a decrease in this activity. A small decrease has previously been found at the motor endplates of the gastrocnemius muscle of guinea-pigs after denervation, whereas in the superior cervical ganglion of cats a considerable reduction of cholinesterase concentration was observed after section of preganglionic fibers. It was suggested that the amount of reduction depends upon the surface extent of the degenerated nerve fibers. Therefore, the difference between endplates and ganglia may be partly attributed to the extensive endarborization of preganglionic fibers not paralleled by the motor nerve endings in guinea-pig muscles.

The effect of nervous degeneration upon cholinesterase activity within the central nervous system has now been studied. Unilateral section of the dorsal roots of the IIId to VIIth lumbar segments of the spinal cord was carried out in cats, and the enzyme concentration determined in the dorsal and ventral horns of the VIth lumbar segment at various intervals after operation.

In normal unoperated cats the average Q CH.E. (mgm. acetylcholine split by 100 mgm. of fresh tissue in 60 min.) were 16.1 and 16.0 for the left and right dorsal horns respectively. In the ventral horns the average Q CH.E. values were 20.4 (left) and 21.7 (right). In the operated cats the cholinesterase concentration decreased approximately equally in all four horns. The average Q CH.E. fell to 10.9 (minus 32 per cent) in the left and 11.7 (minus 27 per cent) in the right dorsal horn. For the ventral horns the corresponding values were left 14.7 (minus 28 per cent) and right 16.3 (minus 25 per cent). After 6 days the maximum reduction of Q CH.E. has already been reached and there is no further reduction in animals sacrificed as late as 40 days after operation. It is noteworthy that the period of maximal decrease in Q CH.E coincides with the period during which the boutons terminaux undergo degeneration.

The cause of death in experimental anuria.[2] H. E. HOFF, P. K. SMITH (by invitation) and A. W. WINKLER. *Laboratories of Physiology and Pharmacology, and Department of Internal Medicine, Yale University School of Medicine, New Haven, Conn.*

Dogs in which the kidneys have been aseptically removed or the ureters ligated were followed until death. Electrocardiograms and blood samples were obtained at intervals before and at the exact moment of death. Concentration of potassium in serum rose progressively until a level capable in itself of causing death was attained. The sequence of electrocardiographic changes prior to death was identical with the quite characteristic

[1] Aided by a grant from the Dazian Foundation.
[2] Aided by grants from the Committee on Therapeutic Research of the American Medical Association, the Ella Sachs Plotz Fund and the Fluid Research Fund, Yale University School of Medicine.

sequence in normal animals associated with continuously rising concentration of potassium. These include: first, an increased height of the T wave followed by a drop in the ST segment; then disappearance of the P wave, and, finally, intraventricular block and cardiac arrest. Administration of potassium salts in small amounts immediately following the initial establishment of anuria was followed by death at an earlier time and with less increase of the blood nonprotein nitrogen, but with the same level of potassium in serum. It is concluded that death in this type of anuria is due to cardiac arrest induced by an elevated concentration of potassium in serum, which is in turn due to a breakdown of tissue and failure of the body to excrete or store released potassium.

Dogs with anuria following intravenous injection of mercury usually died from some other cause before the concentration of potassium had risen sufficiently to cause death. Potassium poisoning is not the usual cause of death in human nephritis.

Resistance to slowly increasing doses of sodium pentobarbital[1] in the white rat: duration of higher tolerance after parturition and effects of age, sex, castration and administration of testosterone propionate.[2] HARALD G. O. HOLCK[3] and DONALD R. MATHIESON (by invitation). *Department of Physiology and Pharmacology, College of Pharmacy, University of Nebraska, Lincoln.*

Development of tolerance and ability to destroy pentobarbital were studied in two experimental series upon 536 rats divided into 25 groups. The starting subcutaneous dose of sodium pentobarbital was 11.7 or 10.9 mgm./kgm. and every 90 minutes another injection was made until each animal died; successive dose increases were 3.22 or 4.5 per cent. Chief results: I. *Postpartum:* The high percentage of tolerance observed three weeks after parturition was absent five weeks later. II. *Age and sex:* Rats (both sexes) one month old were most resistant to pentobarbital; those two months old—particularly the females—distinctly less, and full grown rats least of all, again the females less than the males. III. *Castration:* In adult males castration reduced the tendency to develop tolerance but not the ability to destroy pentobarbital once tolerance was established. Thirty-five per cent of the spayed females became tolerant, only 9 per cent of the unspayed animals; the tolerant spayed females, when two months old, destroyed more pentobarbital than the unspayed (97 against 74 mgm./kgm.). IV. *Testosterone propionate* (Perandren) Effects: Subcutaneous injections of the oil solution of the ester equivalent to 1 mgm./kgm. of testosterone (2 mgm. in one series of normal males and females) were made daily for 3 or 10 full days prior to and daily during the pentobarbital injections until death. A. Normal adult males showed no effects. B. Three-day administration increased the number of castrated males developing tolerance (83 against 52 per cent). The 10-day administration was still more effective (95 per cent). C. Sixty-five per cent of the normal adult females receiving hormone 10 days developed tolerance as compared with 9 per cent of the untreated animals. Three-day administration pro-

[1] Courtesy of Eli Lilly and Company.

[2] Donated by Ciba Pharmaceutical Products.

[3] Aided by a grant from the Committee on Therapeutic Research, Council on Pharmacy and Chemistry, American Medical Association.

duced no significant change. Two mgm./kgm. were not more effective than 1. D. The percentage of spayed females developing tolerance was markedly increased by the 10-day and less by the 3-day administration (85 and 74, respectively, compared with 35 per cent in untreated castrates). Capacity to destroy pentobarbital was increased in the tolerant spayed females after 10-day hormone treatment.

The synchronization of cerebro-cortical potentials. C. G. HOLLAND (introduced by S. W. Britton). *Physiological Laboratory, University of Virginia Medical School, University.* (Read by title.)

Alpha and delta activities as recorded by the electroencephalograph represent energy output per unit time interval. This energy is variable, depending on the activity of cortical neurones during the interval measured.

In 20 normal subjects and in 10 cases representing various pathological states it was found, by direct measurement of the tracings, that there was a parallelism between delta and alpha activity for total outputs of energy from second to second. The same parallelism was, moreover, apparent when alpha and delta activities were reduced to average waves and compared.

In this latter case a proportional correspondence was evident. Thus, for example, when in a certain subject the average duration of delta waves (produced by hyperventilation) was found to be 4 times the duration of the average normal alpha deflection, the average peak output for delta waves was approximately 4 times that of the average alpha deflection.

It is suggested therefore that delta deflections, whether occurring in normal or pathological subjects, represent summations of normal alpha activity.

The effect of positive and negative intra-thoracic pressure on right auricular and peripheral venous pressure. J. P HOLT (introduced by Hampden Lawson). *Department of Physiology, University of Louisville School of Medicine, Louisville, Ky.*

It has been shown (Lyons et al. Am. Heart J. **16:** 675, 1938; Holt. Am. J. Physiol. **130:** 635, 1940) that the collapse of peripheral veins may maintain the peripheral venous pressure at a normal or high level. Since it is generally agreed that the pressure in the right auricle is sub-atmospheric and that the pressure in the veins just before entering the chest is sub-atmospheric, it was thought that small changes in right auricular pressure might not affect peripheral venous pressure because the veins just before entering the chest might be partially collapsed.

In barbitalized dogs, with the chest closed, and placed in the supine position, peripheral venous pressure was measured in the femoral, cephalic, or jugular vein by a modification of the method of Moritz and Tabora. Right auricular pressure was measured by means of a water manometer connected to a cannula that passed into the right auricle by way of the external jugular vein. The trachea was cannulated and connected to a breathing chamber in which the pressure was varied between 20 cm. above and 20 cm. below atmospheric pressure.

The auricular pressure decreased markedly when negative pressure was applied in the chamber, while peripheral venous pressure remained approximately constant. When small positive pressures were applied in

the chamber auricular pressure increased while in some cases peripheral venous pressure remained constant. When high positive pressures were applied in the chamber both auricular and peripheral venous pressures increased. The veins which enter the upper end of the chest were observed normally to be partially collapsed and to collapse further with negative pressure in the chamber. Similar results were obtained on models using thin walled collapsible tubes to represent veins.

It is suggested that peripheral venous pressure is maintained at its normal value when auricular pressure is decreased and, in some cases, when auricular pressure is increased, because the veins just before entering the chest are collapsed to a greater or lesser degree depending on the auricular pressure, and that the collapse changes the resistance to the flow of blood along the veins and maintains peripheral venous pressure approximately constant.

The respiration of brown adipose tissue and kidney of the hibernating and non-hibernating ground squirrel. WALTER E. HOOK and E. S. GUZMAN BARRON (introduced by A. J. Carlson). *The Lasker Foundation for Medical Research and the Department of Medicine, University of Chicago, Chicago, Ill.*

The brown adipose tissue of the ground squirrel shows considerable metabolic activity when compared with white adipose tissue and with tissues of high metabolic activity. The O_2 consumption of slices of brown adipose tissue was 17.1 ± 3.65 c. mm. per mgm. of dry, fat-free weight. The optimum activity was found at pH 7.31 with O_2 as gas phase. The respiration seemed to show seasonal variation with the lowest figures in summer and the highest in fall. The R.Q. was 0.80. The anaerobic glycolysis in the absence of glucose was 4.0 c.mm. CO_2; on addition of glucose it rose to 6.0. The tissue oxidized succinate and pyruvate with the same activity as kidney; it also oxidized lactate, citrate, α-ketoglutarate, fatty acids, and amino acids. The respiration was almost completely abolished by HCN. The tissue contained cytochrome c, and the activity of its cytochrome oxidase was 14 per cent that of the heart. The diphosphothiamine content of the tissue was 18 micrograms per gram, three times that of the liver, and decreased only 15 per cent after six weeks hibernation. The ascorbic acid content of the tissue was 0.111 mgm. per gram, one-third that of the liver. The respiration of the kidney at 8°C. was 15 per cent that of the respiration at 38°C., whereas the respiration of brown adipose tissue was 36 per cent. In hibernation, therefore, while other tissues reduce their metabolism to a minimum, brown adipose tissue still retains one-third of its optimum activity.

Effect of lowered body temperature on heart rate, blood pressure, and electrocardiogram. W. E. HOOK and R. T. STORMONT (introduced by E. M. K. Geiling). *Departments of Medicine and Pharmacology and the Otho S. A. Sprague Memorial Institute, University of Chicago, Chicago, Ill.* (Read by title.)

The body temperature of dogs and cats anesthetized with ether or sodium barbital was reduced by the application of crushed ice to the animal's body. Deep rectal temperatures were observed. The blood pressure

was recorded manometrically from the carotid artery in the cats and was taken by repeated femoral puncture in the dogs. No essential differences were noted between the two species. Electrocardiograms in the dogs were taken with the three conventional leads. The averages of the results obtained are summarized in the following table:

Body temperature, °C.	38	34	30	26	22	18
Heart rate	163	141	114	74	50	22
Blood pressure (mm. Hg)	128	116	98	93	56	31
P-R interval (sec.)	0.10		0.13		0.22	
QRS complex (sec.)	0.07		0.08		0.11	

Electrocardiograms during progressive reduction of body temperature showed gradual development of a marked sinus bradycardia with prolongation of the P-R interval, QRS complex (with notchings), and QT segment. In addition other changes not ordinarily associated with a slow rate were seen in the ST segment and in the T wave which became diphasic and then deeply and bizarrely inverted as the body temperature approached 20°C. Electrocardiograms taken immediately after recovery to normal body temperature in three animals which had been cooled to 19°C. showed no significant changes from normal.

As the body temperature was reduced the amount of ether required to maintain anesthesia was gradually decreased until at 22°–20°C. ether administration could be discontinued. Below 20°C. artificial respiration was frequently necessary to maintain life. Recovery occured in some animals even when the body temperature had been reduced to as low as 17°C. provided that the animal was warmed by the application of external heat.

Poletop method of artificial respiration. D. R. Hooker, W. B. Kouwenhoven (by invitation) and O. R. Langworthy. *Departments of Physiological Hygiene, Electrical Engineering and Medicine, Johns Hopkins University, Baltimore, Md.* (Motion picture demonstration.)

The method as used in the field to apply artificial respiration to men injured by contact with live power lines is demonstrated. It consists of manual application of abdominal compression to the subject in the vertical position and has the advantage of being applicable within a brief period after the accident.

Tests were run on conscious subjects to compare the volume of air moved by this method and by the Schaefer method. The results obtained indicate that the new method is of equal if not greater value under the conditions established. No effort has been made to test the value of the method for subjects on the ground.

Average results on fifteen individuals were:

	CONTROL RATE PER MIN.	CONTROL VOL. PER RESP.	TIME TO MOVE UNIT VOL. AIR	EXPER. RATE PER MIN.	EXPER. VOL. PER RESP.	TIME TO MOVE UNIT VOL. AIR
Schaefer prone	13.2	546	*1′ 22″*	10.8	1028	*54″*
Poletop upright	14.9	558	*1′ 12″*	10.3	1363	*43″*

Possible rôle of the kidney in the maintenance of normal blood pressure.[1]

R. M. House (by invitation) and G. E. Wakerlin. *Department of Physiology, College of Medicine, University of Illinois, Chicago.* (Read by title.)

The effect of bilateral splanchnicotomy on the blood pressures of twelve bilaterally nephrectomized and twelve dummy nephrectomized dogs was studied. The decreases in blood pressure in the experimental and control groups following splanchnicotomy were variable but not significantly different. The effect of cordotomy at the level of the sixth thoracic segment on the blood pressure of nine bilaterally nephrectomized and nine dummy nephrectomized dogs was studied. The reduction in blood pressure in both the nephrectomized and the dummy nephrectomized dogs following cordotomy was variable but not significantly different. Evidence was obtained to support the view that the trauma associated with exposing the spinal cord is more responsible for the fall in blood pressure previously reported in the physiological literature than is the sectioning of the spinal cord at the sixth thoracic segment. These results lend no support to, but by no means rule out, the possibility that the normal kidney plays a rôle in the maintenance of arterial blood pressure in the dog.

The effect of adrenalectomy with desoxy-corticosterone substitution therapy on the seminal vesicles and prostate in castrated mice and rats.

Evelyn Howard. *Department of Physiology, Johns Hopkins School of Medicine, Baltimore, Md.*

It has been suggested that the anomalous post-castration maintenance of columnar epithelium in the seminal vesicles and prostates of young mice and rats might be due to a temporary phase of andromimetic activity on the part of the adrenal cortex. If this hypothesis is correct, and if the life-maintaining adrenal hormone is not androgenic, it should be possible to maintain castrated adrenalectomized animals in good condition on substitution therapy, without obtaining the anomalous columnar epithelium in the vesicles and prostate.

Subcutaneous pellets of desoxy-corticosterone acetate maintain normal growth in young adrenalectomized mice. In castrated adrenalectomized adults carrying pellets it was found that the seminal vesicle epithelium was in a predominantly cuboidal condition, little if at all superior to the state of the untreated castrate. In three weeks old adrenalectomized castrates with pellets, however, the epithelium was maintained in the columnar condition characteristic of castrates at this age, although in untreated animals this columnar state can be abolished by adrenalectomy. It seems apparent that in mice this anomalous maintenance of seminal vesicle epithelium must be due either to intrinsic differences between the immature and the adult castrates in the reactivities of the end organs, or to a transitory source of androgens outside of the gonads and the adrenals.

The ventral prostate degenerates after adrenalectomy in castrated three week old rats, and this degeneration is not prevented by desoxy-corticosterone acetate, according to Burrill and Greene, who did not, however, give body growth data. I have confirmed their observations on rats, and find that castrated adrenalectomized animals maintained on desoxy-

[1] This work was aided by a grant from the Graduate School Research Fund of the University of Illinois.

corticosterone acetate pellets with ninety per cent of the body growth of castrated litter mates show a fifty per cent reduction in prostate size compared to the castrates, but no marked differences in size or histological state compared to untreated castrated adrenalectomized litter mates, with thirty-four per cent body growth. The histological state of the glands in castrates is superior to that in adrenalectomized castrates. It may be concluded that in young rats there is an appreciable temporary andromimetic activity of the adrenal cortex.

Production of glycosuria in the normal rat by stilbestrol and by 17-hydroxy-11-dehydro-corticosterone. DWIGHT J. INGLE. *The George S. Cox Medical Research Institute, University of Pennsylvania, Philadelphia.*

It has been previously shown that 17-hydroxy-11-dehydro-corticosterone and stilbestrol are among the substances capable of intensifying the diabetic state of the partially depancreatized rat. The experiments were extended to normal male rats, weighing approximately 300 grams which were maintained on a high carbohydrate diet administered by stomach tube twice daily. The available carbohydrate of the diet was 15 grams daily. When stilbestrol was administered hyperglycemia and glycosuria were induced in over two-thirds of a large series of animals studied. Daily doses as small as 50 gamma were effective. Adaptation occurred so that the diabetic state disappeared after being continually present for periods of 2 to 36 days. Three normal rats were treated with 17-hydroxy-11-dehydro-corticosterone. One animal treated with 5 mg. daily developed hyperglycemia and mild glycosuria, the second, also treated with 5 mg. daily developed hyperglycemia and severe glycosuria, and the third treated with 10 mgm. daily succumbed during hyperglycemia and severe glycosuria. In these experiments there was always an increase in the excretion of non-protein nitrogen during the period of glycosuria but the total protein catabolism thus measured was too small to account for all of the glucose excreted. None of these animals excreted ketones although ketonuria is frequently observed during treatment of the partially depancreatized rat with certain estrogens and adrenal steroids.

The work performance of adrenalectomized rats treated with 11-desoxy-corticosterone sodium phosphate and with 11-desoxy-17-hydroxy-corticosterone. DWIGHT J. INGLE. *The George S. Cox Medical Research Institute, University of Pennsylvania, Philadelphia.* (Read by title.)

Earlier studies have shown that 11-desoxy-corticosterone and its acetate are very weak in their effects on the work performance of adrenalectomized rats as compared to the effects of those compounds which are identical in structure except for the presence of oxygen at carbon 11, or for the presence of oxygen at carbon 11 and at carbon 17. One of the explanations suggested to account for the wide difference in activity is that 11-desoxy-corticosterone is much less soluble than the more highly oxygenated compounds and may not be readily absorbed in acute experiments. Reichstein has prepared the sodium salt of 11-desoxy-corticosterone phosphate which is soluble in water. The effect of this substance on work was not enhanced by its administration in this soluble form. This suggests that the apparent difference in the principal biologic effects of the adrenal steroids is not due to differences in solubility. A second question con-

cerned the influence of the oxygen atom at carbon 11, and at carbon 17, with respect to the effect on work. It was possible to answer this question when Reichstein prepared 11-desoxy-17-hydroxy-corticosterone by partial synthesis. An examination of a sample of this compound kindly supplied by Professor Reichstein showed that the presence of the hydroxyl at carbon 17 had no more effect on work than the weakly active 11-desoxy-corticosterone. Thus it appears that the presence of an oxygen atom at carbon 11 is essential for a favorable effect on work.

The nature and reversibility of some effects of pH changes on erythrocytes. M. H. JACOBS. *Department of Physiology, University of Pennsylvania, Philadelphia.*

In earlier communications attention has been called to the highly characteristic manner in which pH changes affect the apparent permeability to glycerol of the erythrocytes of different species of mammals. These results were obtained by the hemolysis method, which involves several possible complicating factors. Further studies have therefore been made of the rates at which, at different pH values, volume changes of erythrocytes occur when a penetrating solute is suddenly added to a suspension of the cells in a buffered isotonic salt solution, recording the changes by the optical method of Parpart.

On the whole, the results obtained by this method are in good agreement with the earlier ones; but they permit a separation of equilibrium volume factors from permeability factors that is impossible with the hemolysis method. At sufficiently high and low pH values the former are of considerable importance, but, in general, permeability and rate of hemolysis tend to run parallel.

Of special interest is the high degree of reversibility of the effects of pH changes on permeability to glycerol. If to a suspension of erythrocytes of the "human" type, undergoing a slow restoration of volume in a glycerol-containing salt solution at pH 5.4, sufficient alkali be added to raise the pH to 7.4, the characteristically rapid rate of entrance of glycerol into the cells, and of swelling of the latter, immediately returns. This may, in turn, be almost instantly checked by the addition of more acid, and again restored by the addition of alkali, the recovery curve thus becoming a series of steps corresponding to the alternate additions of acid and alkali. Furthermore, if, after complete restoration of volume has occurred at one pH value, acid or alkali be added, a new curve, characteristic of the changed pH, can be obtained on the addition of more glycerol. Such changes may be repeated many times.

That these effects are related specifically to the permeability of the cells to glycerol is suggested by the fact that they have not as yet been obtained with other penetrating solutes, except to a much lesser extent with ethylene glycol.

Contraction potentials (Quadriceps femoris) in man during reading. EDMUND JACOBSON and FRANCES L. KRAFT (by invitation). *Laboratory for clinical Physiology, Chicago, and Wells College.*

Action-potentials were measured in unselected individuals while seated, reading. They were 39 males and 61 females ranging in age between 22 and 46 years, mostly employees in various business occupations. Fine

platinum, irridium needles were used as electrodes, inserted above the patella in the midline of the thigh two or more inches from each other. The left foot rested on a support while the right foot was free. An a.c. amplifier and Integrating Microvoltmeter were employed. The rectified action-potentials were averaged and integrated every two minutes during a thirty minute period of test. The results can be presented 1, as averages of the potential differences for the entire thirty minute period; 2, as curves of the integrated action-potentials plotted against time.

The distribution curve (plotted on a 0.5 microvolt scale) of the averaged potentials for the entire group is of the normal variety, ranging from 0.5 to 4.5 microvolts with the maximum number of individuals (35) in the range between 1.0–1.5 microvolts.

Under the conditions of test, the course of the composite curve for 100 individuals over the thirty minute period runs at all points between the values of 1.5 and 2.0 microvolts. The highest levels occur approximately from 0–2 and 26–30 minutes, while the lowest level occurs from 16–18 minutes.

The curves are (approximately) descending in 26 instances, (approximately) ascending in 41 instances and (approximately) horizontal in 33 instances. As interpreted in the light of previous investigations on the knee-jerk, in this group of apparently healthy individuals, a distinct trend toward differential relaxation while reading is not exhibited.

Electrical oscillations from insect eyes. THEODORE L. JAHN and FREDERICK CRESCITELLI (introduced by J. H. Bodine). *Departments of Zoology, University of Iowa, Iowa City, and of Physiology, University of Washington, Seattle.*

Electrical oscillations (8–60 cycles per second; 10–100 microvolts) have been recorded from the compound eyes of five species of grasshoppers, four species of moths, and two species of butterflies.

In grasshoppers certain types of oscillations are found to be associated with continuous illumination of the dark adapted, light adapted, and intermediate adapted eyes, and these oscillations are different for the various conditions of adaptation. Since they occur during illumination they are referred to as "light" rhythms. Another type of light rhythm occurs only with the "on" effect of continuous illumination of the dark adapted eye, and a different rhythm is also present after cessation of prolonged continuous illumination. These two types are referred to as "on" and "after" rhythms. The influence of temperature and of the intensity of light on the frequency and amplitude of the intermediate adaptation rhythm was studied. Another type of after rhythm is found only in response to brief exposures of the dark adapted eye to high temperatures.

In moths there are both light and after rhythms to long exposures, and there are two after rhythms of quite different frequencies to short exposures. In the latter cases the amount of light rather than intensity or exposure determines the nature of the rhythm. A strong stimulus elicits a high frequency (25–30 per sec.); a weak stimulus elicits a low frequency (8–10 per sec.); and intermediate stimuli may elicit both frequencies.

In the dark adapted butterfly response there is a light rhythm and an after rhythm, neither of which is recorded from the light adapted eye.

Removal of the proto-, deuto-, and tritocerebral hemispheres did not

affect the character of the rhythms in the grasshopper. It is assumed that they originate in the optic ganglion.

An attempt will be made to interpret these results in terms of synchronization of ganglionic neurone responses, and in terms of the various types of fibers known to occur in the optic nerves of other animals.

The origin of the electrical response obtained from the compound eyes of grasshoppers. THEODORE L. JAHN and V. J. WULFF (introduced by J. H. Bodine). *Department of Zoology, State University of Iowa, Iowa City.* (Read by title.)

The electroretinogram of the vertebrate eye is usually considered to originate in the ganglionic neurones of the retina. Within recent years it has been demonstrated that the a-, b-, c-, and d-waves characteristic of the vertebrate electroretinogram also occur in the electrical response of the compound eyes of insects. It has been stated that the origin of this electrical change is probably in the optic ganglion, but experimental evidence has been lacking.

Present experiments on the grasshopper Trimerotropis maritima have demonstrated that the characteristic response of the dark adapted eye (consisting of b- and c-waves) is not changed appreciably by excision of the brain and optic ganglia. These phases of the electrical response, therefore, must originate within the eye. In some experiments in which the back of the eye was damaged during removal of the ganglion the c-wave was no longer present. After light adaptation for several seconds the b-wave disappeared, and the response became a slow wave in the direction opposite from that of the normal c-wave. This change in the response is similar to that obtained in vertebrate eyes with injury, low temperature, or deep anaesthesia.

In addition to the a-, b-, c-, and d-waves the response of the grasshopper eye may contain several types of electrical oscillations. The effect of excision of the optic ganglion on two types of these oscillations (intermediate adaptation rhythm, and high temperature after rhythm) was studied. In no case was it possible to record these rhythms after excision of the ganglia. It is concluded that the rhythms probably originate in the optic ganglion.

Hydrodynamic factors determining pulse pressure.[1] KENNETH JOCHIM (introduced by L. N. Katz). *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.*

This study was undertaken in order to evaluate the various factors determining the magnitude of the pulse pressure in an elastic system with pulsating fluid flow. Since the circulatory system of an animal contains many variables that are not amenable to mathematical analysis, an artificial system was constructed. The pulsating flow is supplied by a piston type pump, the rate and stroke output of which can be varied at will and accurately controlled; the velocity of output has the form of a sine curve. The elastic portion of the system consists of a length of distensible rubber tubing, an air cushion, or a combination of the two, so that any desired type of pressure—volume curve (which characterizes an elastic system) may be obtained. The rest of the system is completely rigid. A rigid

[1] Aided by the A. D. Nast Fund for Cardiac Research.

arrangement is also provided by means of which the peripheral resistance may be varied within wide limits.

Pressure within the elastic system was recorded with a Hamilton manometer; a velocity pulse was recorded on both sides of the elastic portion with the electromagnetic flowmeter. The volume pulse of the elastic portion was determined by integration of the velocity curve on the distal side of the elastic system.

The first problem studied was the effect on pulse pressure of increasing the peripheral resistance, other factors remaining constant. If the pressure-volume curve of the elastic system used is concave toward the volume axis, the pulse pressure decreases as peripheral resistance increases. If the P-V curve is concave toward the P axis, the pulse pressure increases as the peripheral resistance is increased. If the P-V curve is a straight line, the pulse pressure is constant, regardless of the peripheral resistance. In other words, the magnitude of the pulse pressure (with a fixed pump rate and stroke output) depends on the mean slope of the P-V curve in the working range. Changing the peripheral resistance merely shifts the working range along the P-V curve to another region where the mean slope may be less, greater, or the same.

Antihormone for vasopressin (antivasopressin).[1] C. A. JOHNSON (by invitation) and G. E. WAKERLIN. *Departments of Physiological Chemistry and Physiology, College of Medicine, University of Illinois, Chicago.* (Read by title.)

Six rabbits were given daily intramuscular injections of vasopressin (pitressin) solution (1 cc. = 10 units) in a dosage of approximately 1½ units per kgm. for a period of six weeks. At the end of this period the serums of all these rabbits completely neutralized the acute pressor response to vasopressin when assayed intravenously on the brain-pithed nephrectomized cat. The assay mixture consisted of 1 cc. serum and 0.1 unit vasopressin previously mixed and allowed to remain at 4°C. overnight. The assays were suitably controlled with normal rabbit serum mixtures as well as saline diluted vasopressin. We purpose to study the effect of this pressor neutralizing factor or antihormone on other physiologic and pharmacologic actions of vasopressin.

Production of antirenin by heterologous renins.[1] C. A. JOHNSON (by invitation), G. E. WAKERLIN and M. L. GOLDBERG (by invitation). *Departments of Physiological Chemistry and Physiology, College of Medicine, University of Illinois, Chicago.*

In earlier experiments we reported (Proc. Soc. Exper. Biol. and Med. **44:** 277, 1940) the production of antiserums for dog renin in the rabbit and for hog renin in one dog. These observations have now been extended to 5 dogs given daily intramuscular injections of hog renin for 4 months each. Four of the animals were hypertensive, one normotensive. By the end of the first month, the serums of all of the dogs contained a substance (antirenin) which neutralized the acute pressor response to hog renin when assayed on the nephrectomized etherized dog or on the

[1] This work was aided by a grant from the Graduate School Research Fund of the University of Illinois.

nephrectomized brain-pithed cat. The assay mixtures consisting of 2 parts of antiserum and 1 part renin (equivalent to ½ gram of fresh kidney per kilogram of assay animal) were administered intravenously and properly controlled with normal serum mixtures. The serums of these dogs similarly negated the acute pressor response to dog renin.

Normotensive rabbits injected intrasmucularly or intravenously with hog renin likewise developed a pressor neutralizing substance for the injected renin. The kidneys of these rabbits showed a normal renin content.

Three groups of guinea pigs were injected intraperitoneally with dog, hog, and rabbit renins respectively for periods of 2 to 5 months. Preliminary findings in these animals have also demonstrated the presence of antirenin in the serums.

Two hypertensive dogs injected intramuscularly for 4 months with heat-inactivated hog renin failed to develop antirenin to either hog or dog renin. Likewise, normotensive rabbits injected with heat-inactivated hog and dog renins failed to develop antirenin.

One hypertensive dog treated with active dog (homologous) renin for 4 months showed no evidence of antirenin for either dog or hog renin. This finding strongly suggests that the production of antirenin depends on the heterologous character of the injected renin.

Relation between initial fiber length and force of contraction in the left ventricle. J. RAYMOND JOHNSON and J. R. DIPALMA (by invitation). *Department of Physiology and Pharmacology, Long Island College of Medicine, Brooklyn, N. Y.*

The force of contraction of cardiac muscle should be considered as a direct function of the intramuscular tension developed in the heart and it does not necessarily parallel measurements of cardiac output or of intraventricular pressure. Proceeding on this basis and using the method which we described previously (Am. J. Physiol. **125:** 234, 1939) of recording the intramyocardial pressure pulse from an imbedded artery segment, we have undertaken experiments to determine the effects of increased and decreased diastolic filling on contraction in the left ventricle.

When the venous return was increased by infusion of 30 to 40 cc. of saline into the femoral vein, the intramyocardial pressure pulse showed a drop of 4 to 32 mm. Hg in spite of the fact that there was a simultaneous marked increase in the systolic aortic pressure accompanied by an appreciable increase in aortic pulse pressure. Decreasing the venous return by compression of the inferior vena cava produced effects in the opposite direction. Prolonged compression resulted in a subsequent fall in the intramyocardial pulse accompanied by extremely low aortic pressures, but if the compression was released before the intramyocardial pressure fell below normal levels the effects seen were again those of an increased venous return.

These results still obtain after section of both vagus nerves. They are not produced reflexly by changes in the aortic pressure as shown by experiments in which this pressure was altered by increasing and decreasing arterial resistance. They are therefore believed to be due directly to changes in the initial length of the cardiac muscle fibers.

The following conclusions are reached: 1, aortic pressure changes cannot

be regarded as a criterion of even the qualitative changes in the force of contraction in the left ventricle; 2, increasing the initial length of muscle fibers in the left ventricle through augmented diastolic filling results in a diminution of its force of contraction; decreasing the initial fiber length by reducing the diastolic inflow results in contractions of greater force.

The enterohepatic circulation of bile acids. CHARLES G. JOHNSTON and J. LOGAN IRVIN (by invitation). *Department of Surgery, Wayne University College of Medicine, Detroit, Mich.*

The enterohepatic circulation of bile acids has been studied by the use of hogs prepared by cholecystectomy and insertion of catheter tubes into the proximal and distal ends of the severed common duct.

Cholic acid is present normally in ox but not in hog bile. Therefore, administration of dried ox bile to hogs permits study of enterohepatic circulation by determinations of the cholic acid excreted in the hepatic drainage bile.

When small amounts of dried ox bile are administered to the hogs by duodenal tube, there is a high degree of efficiency in the excretion of cholic acid in the hepatic bile collected from the biliary fistulae. However, when larger amounts are given, the efficiency of the excretion diminishes. For example, when 1.15 millimols of cholic acid (contained in dried ox bile) are given by duodenal tube, about 92 per cent is excreted in the hepatic bile in 11 hours. In contrast, only 62 per cent is excreted when 6.75 millimols of cholic acid are administered, and the total period of excretion is prolonged to 22 hours.

The duration of enterohepatic circulation of bile acids after a single oral or duodenal administration of dried ox bile is studied by connecting the catheters from the proximal and distal ends of the severed common duct. At the end of the circulation period, the two tubes are disconnected, and the quantity of cholic acid remaining in the enterohepatic system is determined by analysis of the hepatic bile drained from the common duct during a collection period. When small amounts of cholic acid are administered intraduodenally, about 87 per cent is recovered after one day of circulation, 74 per cent after two days, 52 per cent after four days, and 32 per cent after six days. When larger amounts of cholic acid are given, the loss during enterohepatic circulation is greater.

These observations suggest the magnitude of bile acid synthesis required by normal animals for replacement of the amounts lost during enterohepatic circulation.

On the formation of gall stones in the human gall bladder. K. K. JONES and MARIE LORENZ (by invitation). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

It has been shown by Jones and Lorenz (Am. J. Physiol. **126:** P548, 1939; **129:** P391, 1940) that the disintegration of gall stones in the gall bladder is effected by the solubility of the cholesterol in the bile fats. In this research it has been found that the cholesterol in normal bile is held in solution as a fatty acid—bile salt complex from which it cannot crystallize or even separate as long as fatty acids, insoluble in water, are present in any quantity. Before crystallization can occur, degradation of these fatty acids to short chain water soluble fatty acids must take place. Ex-

amination of gall bladder bile reveals only one possible source of oxygen in this highly anaerobic fluid. This is the biliverdin which is found in relatively large amounts in some human fistula and autopsy biles. Biliverdin in alkaline solutions is reduced to bilirubin by unsaturated fatty acids, these acids in turn being oxidized. This mechanism may explain the formation of cholesterol stones in an apparently normal gall bladder in the presence of stasis.

The antagonistic action of lipocaic and the pituitary in fat transport. ORMAND C. JULIAN (by invitation), DWIGHT E. CLARK (by invitation), CORNELIUS W. VERMEULEN (by invitation), J. GARROTT ALLEN (by invitation) and LESTER R. DRAGSTEDT. *Department of Surgery, The University of Chicago, Chicago, Ill.*

The previous report by the authors that lipocaic inhibits the fatty infiltration of the liver produced by the administration of the ketogenic hormone of the hypophysis to fasting guinea pigs, has been confirmed. The theoretical possibility that the fatty infiltration of the liver which occurs in insulin-treated depancreatized dogs may be due to unopposed action of the ketogenic hormone of the hypophysis was investigated. The hypophysis was removed from ten normal dogs and after varying intervals of time the pancreas was excised. These animals survived the pancreatectomy from 5 days to 4 months. All the dogs, except one, developed marked fatty infiltration of the liver. The rapidity of onset and the severity of the fatty infiltration in these animals was just as great as in the animals whose hypophysis was intact.

A new cephalic cardio-inhibitory substance. HERMAN KABAT (introduced by M. B. Visscher). *Department of Physiology, University of Minnesota, Minneapolis.*

Arrest of the cephalic circulation in dogs in which the heart had been completely denervated resulted in cardiac acceleration. This acceleration averaged 22 beats per minute, the latent period of the effect was 2–4 minutes and the rate fell to or below the original level within 30 seconds to 1 minute following restoration of blood flow. Similar observations were made utilizing the heart-lung-head preparation.

This effect cannot be accounted for on the basis of sympathetic stimulation resulting from incomplete denervation since the time relations of the acceleration differ markedly from those of sympathin and also because asphyxia resulted only in cardiac inhibition. Changes in blood pressure had no effect on the cardiac response to cephalic anemia. Occlusion of the thoracic aorta caused no change in cardiac rhythm.

To explain the phenomenon, one is led to postulate that there may be produced continuously in the head (presumably in the brain) a substance which passes through the blood stream and inhibits the heart. When cephalic blood flow is arrested, disappearance of the substance after several minutes results in cardiac acceleration. When blood flow is restored, a fresh supply of the substance quickly slows the heart.

Changes in carbon dioxide or other acids cannot account for the effect. Acetylcholine or choline cannot be responsible, since the effect persisted after atropine.

Saline perfused through the internal carotid arteries of the freshly-

killed dog was collected, concentrated and extracted. A systematic search of such perfusates for adenylic acid, which is known to slow the heart in the atropinized dog, was unsuccessful. The characteristic reversible complete heart block, produced in the guinea pig by adenylic acid, was never observed.

An active cardio-inhibitory substance, which is soluble in acetone, alcohol and water and is heat stable, has been isolated from such perfusates. Electrocardiograms show a sinus slowing in the dog heart after atropine, and a sinus slowing in the guinea pig heart following intravenous injection of the substance. The effect is rapidly reversible. Other fractions of the perfusate had no effect on the heart of the atropinized dog.

The esterase activity of different parts of the mammalian central nervous system. IRVING KAPLAN, DAVID J. COHN and FREDERICK REICH (introduced by Heinrich Necheles). *Department of Biochemistry, Michael Reese Hospital, Chicago, Ill.*

The triacetin, tributyrin, and ethyl butyrate hydrolyzing activities of different parts of the central nervous system of man, rhesus monkey, dog, rabbit and rat were found to differ with the location of the tissue. Minced fresh tissue was used as the enzymic material. The activity was determined by measuring the number of milliliters of 0.05 N. sodium hydroxide required to neutralize the acid formed per gram of tissue by the breakdown of substrate during a 4 hour incubation period in the presence of 0.05 molar phosphate buffer. In all cases the activity was greatest in grey matter, least in white matter, and intermediate in mixed tissues. The average values found for human brain with triacetin, tributyrin, and ethyl butyrate as substrate were respectively: frontal cortex, 24.5, 13.3, and 5.9; parietal cortex, 25.9, 16.2, and 6.3; occipital cortex, 25.2, 14.4, and 5.6; cerebellar cortex, 28.6, 17.1, and 5.6; caudate nucleus, 31.4, 15.3, and 7.1; mixed cerebral tissue, 23.0, 14.4, and 4.9; mixed cerebellar tissue, 29.0, 14.4, and 4.9; thalamus, 24.0, 12.2, and 5.7; midbrain, 22.0, 9.0, and 4.9; pons, 21.2, 8.1, and 4.1; medulla oblongata, 19.5, 8.3, and 4.3; cervical spinal cord, 15.9 (triacetin); white matter, 14.6, 7.1, and 3.7. Values for the rhesus monkey, dog, and rabbit and rat were similar to those obtained with human brain, indicating that the ester hydrolyzing properties of the mammalian brain are practically independent of species differences. This suggests that the ester splitting activity is associated with basic anatomic or physiologic properties of the nervous system which are common to the different species.

Effect of gelatin upon muscular work in man. PETER V. KARPOVICH and K. PESTRECOV (introduced by Edward C. Schneider). *Department of Physiology, Springfield College, Springfield, Mass.*

1. Five series of tests were performed on 76 persons, two series with controlled diet: 1. Twelve jail inmates, bicycle ergometers work; 2. Thirty campers, swimming (1 and 2 series-controlled diets); 3. Twelve heavy-weight lifters; 4. Six wall-weight pullers; 5. Sixteen college students, bicycle ergometers work. In all groups sham feeding was used, so that the men never knew when they received gelatin or a substitute. The amount of gelatin given daily reached in some cases 64 grams.

2. The jail inmates, ranging from 18 to 50 years of age, worked 5 days

a week at the rate of from 0.159 to 0.261 H.P. until this rate could no longer be maintained. The duration of the experiment was about 5 months. The percent of improvement reached from 75 to 4420 per cent. Working time reached in two cases over 6 hours with an output of 2,659,800 ft-lbs. in a day. No effect of gelatin upon duration or efficiency of work could be noticed.

3. College students worked five days a week at a rate of from 0.33 up to 0.506 H.P. for 10 weeks. Improvement varied from 49 to 334 per cent. No effect of gelatin feeding was observed. The best man never received any gelatin. He continued to exercise for 9 more weeks. His improvement reached 463 per cent, and the riding time was 7 minutes, 30 seconds.

4. No effect of gelatin upon the performance of swimmers, weight lifters or wall-weight pullers was observed.

5. In general, with equal work output, stronger men improved more than weaker ones.

6. A psychological effect was observed on the jail inmates. On the day when gelatin and substitute were given for the first time, the performance markedly improved within an hour after the ingestion.

7. College term examinations caused a drop in performance of the students.

Elimination of the pars nervosa without eliciting diabetes insipidus.[1]

A. D. KELLER. *Department of Physiology and Pharmacology, University of Alabama, University.*

The selective elimination of the entire pars nervosa plus the immediately adjacent hypothalamic tissue (tuber) has been accomplished in the cat without precipitating either a residual or a latent diabetes insipidus. The presence or absence of a latent diabetes insipidus was tested by feeding desiccated thyroid.

In the dog total hypophysectomy except for a minute fragment of the pars tuberalis, including the entire pars nervosa, has been accomplished without precipitating either a residual or a latent diabetes insipidus. Total hypophysectomy plus a slight infringement upon the adjacent tuberal tissue has been accomplished without precipitating a residual diabetes insipidus although a latent diabetes was present. Further, typical "latent periods" have appeared in the early course of the resulting permanent residual diabetes insipidus in dogs following total hypophysectomy plus mild infringement upon the ventral extent of the hypothalamus, whereas following total hypophysectomy plus considerable encroachment upon the hypothalamus there is a total absence of the "latent period." Thyroxin and anterior pituitary extract was used in the dog in testing for latent diabetic tendencies and in addition metabolic studies run simultaneously verified the metabolic stimulating activity of these substances.

It is evident from the foregoing data that intact pars nervosa tissue is not invariably essential for the maintenance of a normal water exchange under ordinary conditions as well as under conditions of increased metabolic activity. Likewise, assuming that diabetes insipidus is an antidiuretic deficiency phenomenon, and barring the possibility of there being as yet unrecognized factors complicating such experiments as these, the exist-

[1] Aided by a grant from the Rockefeller Foundation.

ence of an extrahypophysial antidiuretic elaborating focus is demonstrated. Further, strong support is tendered the contention of Sato, Trendelenburg and others that the hypothalamus is the site of such an extrahypophysial elaborating focus. It is to be pointed out, however, that experiments such as these give no indication as to the nature of the elaborating process.

Marked variability in tolerance to insulin following apparently homologous hypothalamic lesions in the cat.[1] A. D. KELLER. *Department of Physiology and Pharmacology, University of Alabama, University.* (Read by title.)

Tolerance to insulin has been studied in a limited number of cats in which the hypophysis was severed from direct hypothalamic innervation by one of the following operative procedures: 1, isolation of the ventral portion of the hypothalamus (tuber) from the remaining hypothalamic tissue by a semicircular sweep with a narrow blunt probe, and 2, after first isolating the tuberal tissue as in 1, this tissue was then removed with forceps leaving the adjacent tissue of the hypophysial stalk undisturbed. Tolerance to insulin was determined only after the animals attained the chronic state (two months) and the tolerance level was verified by several tests. At least two weeks was allowed to elapse between successive tests. The animals were maintained from six to twelve months after operation during which time the tolerance level remained constant.

During the acute stage following these procedures such animals are occasionally prone to spontaneous hypoglycemic attacks and accordingly these animals were protected during this period by feeding twice daily and adding sugar to their drinking water. A decreased tolerance to insulin of ten to twenty times was encountered in the majority of the animals, a decrease of forty times occurring in one cat. Yet in other animals, following essentially homologous anatomical lesions as judged at operation and at necropsy, the tolerance remained normal or approached the normal.

Compared with similar studies on dogs it is evident that the tendency to the hypoglycemic state in the chronic animal is more subject to selective precipitation following hypothalamic lesions in the cat than in the dog as indicated by a decreased tolerance to insulin (Proc. Soc. Exper. Biol. and Med. **42:** 837, 1939).

It is not beyond the realm of probability that we may be dealing here with a primary deficit in the insulin antagonizing factor in pituitrin. Indeed on the basis of other data bearing on the subject, *this probability is definitely under suspicion.*

The striking absence of some of the effects of hypophysectomy following in instances drastic hypophysectomy procedures in the dog.[1] A. D. KELLER. *Department of Physiology and Pharmacology, University of Alabama, University.* (Read by title).

Ordinary hypophysectomy has consistently yielded adrenal atrophy, a decreased tolerance to insulin and amelioration of pancreatic diabetes. This has been our experience also in instances where total hypophysectomy has been attempted. In the latter cases in some respects these effects have been more profound than following ordinary hypophysectomy.

[1] Aided by a grant from the Rockefeller Foundation.

Yet in other animals where great care was taken at operation to insure attaining total hypophysectomy, the tolerance to insulin approached the normal, pancreatectomic hyperglycemia was more intense, and death occurred sooner than in normal pancreatectomized dogs, and the adrenals were essentially unatrophied or definitely less so than that encountered following ordinary hypophysectomy.

The hypophysectomy in these particular experiments has been complicated by 1, a minute remnant of mixed hypophysial tissue found in the base of the fossa (less than 1 per cent of the whole), and 2, some infringement upon the hypothalamus.

If these apparently abortive results are to be explained on the basis of the tissue remnant, this tissue must of necessity be extremely potent in its selective protective activity. Further, remnants left in the fossa retain such functional activity only infrequently because sizable portions of the gland, when left in the fossa purposely or otherwise have not prevented the effects alluded to from occurring in the presence of ordinary hypophysectomy. In two animals exhibiting abortive results no accessory hypophysial tissue was found in the serial sections of the bony sella and underlying tissue.

The only alternative explanation on the basis of our now rather sizable collection of data is that the effects under discussion are *reversed* when, in addition to hypophysectomy, the *environs of the hypothalamus* are encroached upon.

Regardless of the ultimate explanation, the investigation in its present form *emphasizes the rigid necessity* of establishing the "hypophysectomized state" in the dog by comprehensive quantitative functional and necropsy criteria. *Careful operative inspection*, even under excellent direct visibility, *gross and microscopic examination of the surgical specimen* and serial section of the brain base and *contents of the sella turcica* at necropsy, are not by themselves adequate criteria.

Directional course of the axons of the substantia nigra cells as indicated by retrograde degeneration of these cells.[1] A. D. KELLER and L. E. HARRIS (by invitation). *Department of Physiology and Pharmacology, University of Alabama, University.* (Read by title.)

The cells of the substantia nigra disappear in the course of time homolaterally following hemisection and bilaterally following 1, transection of the brainstem through the cephalic midbrain or caudal diencephalon, and 2, a section which more or less selectively demarcates the caudal aspects of the hypothalamus. Hemisection of the brainstem through the middle of the midbrain results in degeneration of the cells caudad to the section while those cephalad to it remain intact. Hemisection or transection of the brainstem at or below the caudal aspects of the midbrain causes no retrograde degeneration of these cells.

Providing we are not dealing with transneural degeneration it is concluded that the substantia nigra cells discharge their axons cephalad and that they course at least as far as the hypothalamus. The ultimate site of the termination of these axons is being investigated.

[1] Aided by a grant from the Rockefeller Foundation.

The toxic effects of intravenously injected calcium solutions. A. B. KENDRICK (by invitation), PAUL BEDINGER (by invitation) and ROBERT W. KEETON. *Department of Medicine, University of Illinois College of Medicine, Chicago.*

It is well known that calcium solutions injected intravenously may at times prove toxic. The necessity for their clinical use demands that this toxic action be well understood.

In experiment A, dogs were injected at a uniform rate with 8.4 per cent calcium gluconate until death occurred. The changes occurring in the electrocardiographic and blood pressure tracings and the relationship of death to serum calcium levels will be discussed. In animals surviving a longer period of injection the percentage of serum proteins was markedly decreased. There were no differences noted between normal and parathyroidectomized animals.

In experiment B a single injection of a toxic dose of either calcium gluconate or calcium chloride was administered. The plasma volume, serum proteins, hematocrit, hemoglobin and serum calcium were determined before and after the injection. In seven animals dying shortly after the experiment was completed, there was a decrease in plasma volume and total circulating proteins. There was an increase in the volume of circulating cells, the hematocrit, and the hemoglobin values. The concentration of the blood at times was so great as to cause difficulty in taking samples from the veins. At death thromboses were noted in lungs and spleen. On sectioning of the liver there was no free bleeding. In five animals living somewhat longer times these changes in blood volume did not occur. Calcium may cause death by direct action on the heart or by blood volume changes. In the latter case death is doubtless due to the concentration of the blood with its increased viscosity.

Changes in electroencephalograms appearing coincident with growth in infant monkeys.[1] MARGARET A. KENNARD and LESLIE F. NIMS. *Laboratory of Physiology, Yale University School of Medicine, New Haven, Conn.*

Electroencephalographic records taken from 28 monkeys (*Macaca mulatta*) at intervals from the first day of life showed consistent changes with growth. During the first 5 days of life there was little electrical activity. It developed gradually thereafter and simultaneously in all leads. Waves of low amplitude and of a frequency of 2 to 3 per second began to appear during the 2nd and 3rd weeks. During the 2nd month both amplitude and frequency increased, the latter to 4 to 5 per second. Greater variability of pattern became evident with bursts of fine waves alternating with slow. At 6 to 8 months amplitude had increased further, but frequency of slow waves remained 4 to 5 per second.

Two animals one year of age had frequencies of 7 to 8 per second and the patterns recorded were characteristic of those found in older animals. In the more mature animals weighing 2.5 to 6.0 kgm. no further developmental changes appeared. Their records showed greater variations of pattern than those of younger animals. Slow wave frequencies were 7 to 8 per second.

[1] Aided by a grant from Child Neurology Research of the Friedsam Foundation.

There is some correlation between the development and elaboration of cortical potentials and the development of coördinate motor activity. Appearance of cortical potentials during the 2nd and 3rd week of life is coincident with beginning of complex "voluntary" movement. A six-months infant can perform all motor acts of an adult animal, but co-ordination, speed and skill develop noticeably during the second 6 months.

Experimental cortical ablations from young monkeys have shown, during the first 6 months, at least, a certain non-specificity of function in cortical motor areas in contrast to older animals, which permits greater reorganization of function in the remaining areas of the young. Evidence therefore from cortical potentials, from behavior and from ablation experiments indicates that in the infant *Macaca mulatta* there are developmental changes in the cerebral cortex which take place at least throughout the first year of life.

High vitamin supplementation (B_1, nicotinic acid and C) and the response to intensive exercise in U. S. Army infantrymen.[1] ANCEL KEYS and AUSTIN F HENSCHEL (by invitation). *Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis.*

Eight representative infantrymen were studied for 11 weeks each under closely controlled conditions on standard Fort Snelling Army rations. Analysis of these rations showed them to be good but not remarkable, in terms of hitherto accepted standards, and average, being in neither best nor poorest classes, compared with other U. S. Army Posts. Twice weekly each man marched, with pack and rifle (29 lbs.) at 3.85 m.p.h. on a motor-driven treadmill at 12.5 per cent grade for 15 minutes. Total work was 16,050 to 17,970 kgm. in 15 minutes The temperature was 78°F., humidity 45–50 per cent; other conditions were constant. Three weeks' training were followed by 4 weeks in which 4 men received placebos and the other 4 received daily supplements of 100 mgm. nicotinic acid amide, 5 mgm. thiamin chloride and 100 mgm. ascorbic acid. This was followed by 4 weeks in which the men previously getting placebos received supplements and vice versa. Observations included heart rate, diastolic and systolic sizes and stroke output, blood hemoglobin, lactate and sugar, blood pressure and subjective reports. Only slight training effects occurred.

No evidence was obtained for a favorable effect of this vitamin supplementation when individual records were studied or when groups were compared. At 5 minutes after exercise stroke output averaged, on supplement, 98.9 per cent and systolic heart size 98.8 per cent of the pre-exercise level; on placebos the averages were 98.4 per cent and 97.4 per cent, respectively. The average heart rate in the second 30 seconds following exercise was 147.6 on supplement and 148.1 on placebos. Average maximum lactate was 33.23 mgm. per cent on supplement and 33.17 mgm. per cent on placebos, with average removal rates in recovery 0.0680 and 0.0745 respectively. Two minutes after exercise the average blood sugar was 86.0 mgm. per cent on supplement and 80.8 mgm. per cent on placebos; eight minutes later these values were 84.8 and 85.1 mgm. per cent, respectively. One man, while on placebos, reported marked

[1] Work sponsored by the Surgeon General, U. S. Army, and the National Research Council.

subjective improvement but this was unsubstantiated by the objective measurements. Other subjective reports of slight improvement occurred equally in placebo and supplement periods.

Laboratory apparatus. MICHAEL KNIAZUK (introduced by Hans Molitor). *Merck Institute for Therapeutic Research, Rahway, N. J.* (Demonstration.)

I. *A continuous heart rate recorder.* The above apparatus was designed to record the heart rate of an animal during an acute experiment. The equipment is simple to use, it does not interfere with the other measurements, and the record may be reproduced on any kymograph. The principle is based on amplifying the heart potential with a portable A.C. operated amplifier and feeding the amplified impulses to a mechanical counter-recorder. The latter is separated from the amplifier and can be mounted near the kymograph.

The counter consists of two electromagnetic step motors, that alternate automatically, each counting for a period of ten seconds. The total number of impulses received during this interval is recorded as a proportional linear displacement.

The ten second counting period was chosen as it is of sufficient length to average the heart rate, and short enough to indicate physiological changes of short duration.

II. *Diuresis recorder.* The instrument is a compact, self-contained volume recorder for liquids, adaptable to total volumes ranging from 25 to 500 cc.

The apparatus operates as follows: A movable platinum contact driven by a reversible motor dips into the fluid-collecting chamber. If the liquid in the chamber rises to the platinum points, the circuit of a sensitive electronic relay is closed. The latter starts the reversible motor which raises the platinum points until the circuit is again broken. An ink-writing point coupled to the same motor, records the displacement on the kymograph.

A unitary analysis of afferent vagal fibers stimulated by changes in lung volume. G. C. KNOWLTON and M. G. LARRABEE. *Department of Physiology and Biophysics, Cornell University Medical College, New York City.*

These experiments were designed to furnish data for a classification of the afferent vagal nerve fibers which are stimulated by changes in the volume of air in the lungs. Cats anesthetized with Dial were used. To facilitate controlled changes in the volume of air in the lungs the chest wall was removed and the animal maintained on artificial respiration. One vagus nerve was cut in the neck and arranged for recording the action potentials of single afferent fibers. Inflations and deflations of the lungs were started with the lungs in the expiratory position.

One group of fibers was stimulated only on inflation, showed rapid adaptation, and had a high threshold.

A second group of fibers was stimulated only on inflation, showed slow adaptation and had a low threshold.

A third group of fibers was stimulated by both inflation and deflation, showed rapid adaptation and had a low threshold

A fourth group of fibers was stimulated by both inflation and deflation, showed slow adaptation and low threshold.

No fibers were found which were excited only by deflation.

A comparison of the distinguishing characteristics of these different fiber groups with the characteristics of respiratory reflexes known to be mediated through the vagus suggests that they subserve different reflex functions. For example, it is known that moderate inflation of the lungs gives a well sustained inhibition of respiration. The low threshold, slowly adapting fibers of group two give a similarly maintained response to the same stimulus. We therefore suggest that their activity is inhibitory to inspiration. Larger inflations of the lungs result in a short-lasting inspiratory effort which parallels the activity initiated in the rapidly adapting fibers of groups one and three. These we therefore assume to be excitors of the inspiratory center. The fibers of group three would then also be responsible for the excitation of inspiration known to be brought about by forced deflation of the lungs. We can suggest no respiratory rôle for the fibers of group four.

Experiments on the conduction of sound through the air of the middle ear cavity. H. G. KOBRAK (introduced by A. J. Carlson). *Division of Otolaryngology, The University of Chicago, Chicago, Ill.*

The conduction of sound through the air of the middle ear cavity (non-ossicular air conduction) was studied in experiments on rabbits. A fistula was made into the middle ear cavity and sound energy conducted through the fistula. The function of the cochlea was measured by the acoustic reflex of the middle ear muscles. There was no difference whether the fistula was open or closed as long as tones of threshold intensity were applied. In over threshold experiments the animal showed stronger cochlear function when the fistula was open. It is assumed that the relative importance of the pathways of sound varies with the intensity level. At threshold level the opening of the additional (fistula) non-ossicular air pathway is unimportant. At higher intensity levels the direct air conduction is an appreciable pathway of sound energy. Curves obtained on animals with experimental deafness are demonstrated. It is assumed that surgical fistulae made in deaf patients in a similar way to those in animal experiments cannot be evaluated by threshold hearing tests.

The effect of testosterone propionate on the ash content of the femurs of castrate mice. CHARLES D. KOCHAKIAN and THEODORE G. MARTENS (introduced by John R. Murlin). *University of Rochester, Rochester, N. Y.*

Male mice were castrated at approximately 5 weeks of age and 30 days later were implanted subcutaneously with testosterone propionate (Perandren) pellets. After 60 to 96 days of treatment, the femurs of these animals showed an average increase in ash of 2.2 per cent over those of their castrate controls. This increase was statistically significant.

The calcium phosphorous ratio of the castrates (2.11) was greater than that of the treated mice (2.03). This difference was due to a greater percentage of calcium. The per cent phosphorous was the same for both groups.

One group of animals had completely absorbed their implanted androgens before autopsy and had reverted to the castrate state. The ash contents of the femurs of these animals was similar to those of the castrate mice, but the calcium phosphorous ratio was the same as that of the treated animals.

The in vitro synthesis of carbohydrate by liver slices and diaphragm of normal, adrenalectomized and adrenal cortical extract treated rats. G. F. KOEPF (by invitation), H. W. HORN (by invitation), C. L. GEMMILL and G. W. THORN. *Chemical Division, Medical Clinic, Johns Hopkins University and Hospital and Department of Physiology, Johns Hopkins University, School of Medicine, Baltimore, Md.*

Since removal of the adrenal glands alters carbohydrate metabolism, a study was undertaken of the synthesis of total carbohydrate from non-carbohydrate sources by liver slices of *a*, normal; *b*, adrenalectomized, and *c*, adrenal cotical extract treated rats. The total carbohydrate content of liver slices was measured before and after $2\frac{1}{4}$ hours of shaking in oxygenated Ringer's solution at 37.5°C. In addition, determinations were made of the ability of the diaphragm to form glycogen from glucose. The adrenalectomized rats were maintained in good condition with sodium chloride and used 6–8 days after bilateral adrenalectomy.

In the liver slices of both normal and adrenalectomized rats a very small but definite quantity of carbohydrate was formed in Ringer's solution without added substrate. Pretreatment of animals with adrenal cortical extract did not alter this finding significantly.

Carbohydrate in appreciable amounts was formed by liver slices of normal and adrenalectomized rats from 0.2 per cent d-lactate or 0.4 per cent pyruvate. Although in a few of the experiments the liver slices of adrenalectomized animals formed subnormal amounts of carbohydrate from these substrates, in most instances the adrenalectomized and normal did not differ significantly. Pretreatment with adrenal cortical extract caused an increase in formation of carbohydrate in liver slices in media of 0.2 per cent d-lactate or 0.4 per cent pyruvate.

Various concentrations of alanine failed to form significant amounts of carbohydrate in liver slices of normal or adrenalectomized animals. However 0.4 per cent glutamate readily increased carbohydrate formation in liver slices of the same animals.

The initial total carbohydrate content of liver slices of adrenalectomized rats fasted 24 hours was found to be much lower than that of normals fasted during a similar period. This finding was so striking that it might be used to confirm the absence of accessory adrenal tissue in adrenalectomized rats.

In a medium of 0.2 per cent glucose the increase in glycogen content of the diaphragm of both normal and adrenalectomized rats was of the same magnitude.

These findings indicate that the synthetic production of carbohydrate from non-carbohydrate sources by liver slices is in some instances diminished and in others not significantly changed by adrenalectomy and pretreatment with large amounts of adrenal cortical extract augments this synthesis.

Specific antagonism between methionine and sulfanilamide in E. coli. HENRY I. KOHN and JEROME S. HARRIS (by invitation). *Departments of Physiology and Pharmacology, Pediatrics, and Biochemistry, Duke University School of Medicine, Durham, N. C.*

Using a strain of *E. coli* capable of growth in a basal medium of inorganic salts and glucose, we have determined the effect of adding to the basal medium each of the naturally occurring amino acids, both in the presence and absence of sulfanilamide. Only methionine, at 0.0001M, can antagonize 0.001M sulfanilamide.

This action is specific for methionine, and is not given by ethionine, homocystine, or sulfamethionine. Ethionine and norleucine inhibit growth and synergize the action of sulfanilamide in our medium, probably by competing with and displacing methionine in the cell, since the addition of small amounts of methionine abolishes these inhibitions.

Sulfamethionine (N-sulfanilyl-methionine) neither antagonizes nor inhibits; and this is also true of the *sulfa-* compounds synthesized by us from alanine, asparagine, beta-alanine, cystine, glycine, leucine, norleucine, norvaline, proline, phenylalanine, valine, and amino-butyric and amino-isobutyric acids.

The metabolism of methionine is obscure. Growth does not occur if the ammonia of the basal medium is replaced by methionine. It is not oxidized, decarboxylated, or hydrolysed by washed suspensions of our strain of *coli* in the Warburg vessel (buffered saline pH 7.2, 37°), though it tends to prevent a decline in respiration (glucose). When growth is initiated by adding ammonia, the increase in respiration is greatly accelerated by the presence of methionine.

Although less active than *p*-amino-benzoate, and incapable of antagonizing high concentrations, methionine must account for some loss of potency of sulfanilamide in media containing protein derivatives, where other amino acids probably enhance its activity. Cozymase is without effect, though it antagonizes sulfapyridine in *Staphylococcus* (West and Coburn); three isomers of amino-nicotinic, nicotinic, and pantothenic acids also are inactive.

Further analysis of the factors determining the temperature coefficients of cellular respiration. IRVIN M. KORR. *Department of Physiology, New York University College of Medicine, New York City.* (Read by title.)

Rubenstein and Gerard (1934) showed that the temperature coefficient of respiration (Q_{10}) of the unfertilized sea urchin egg was approximately twice that of the fertilized egg. In a temperature analysis of the changes in respiration which take place on fertilization the author (Korr, 1937) confirmed the shift in temperature coefficient, finding Q_{10} of the fertilized egg to average around 2.3 and that of the unfertilized egg around 4.1.

That study revealed that the acceleration of oxygen uptake of the unfertilized egg by the addition of a hydrogen-carrier (pyocyanine) lowered the Q_{10}. The amount of the lowering was related to the degree of acceleration. This in turn was related to pyocyanine concentration. At the pyocyanine concentration (0.015 per cent) required to elevate respiration to that of the fertilized egg, the temperature coefficient was identical with that of the latter; at all temperatures unfertilized eggs + 0.015 per cent pyocyanine respired at the same rates as the normal fertilized egg.

It was therefore of interest to observe the alterations in rates and temperature coefficients of egg respiration as the result of suppression of dehydrogenase activity by graded concentrations of ethyl urethane. The fertilized eggs of that series (summer of 1938), observed over a range of temperatures from 12.5° to 30°C., showed a temperature coefficient of 2.5. In the presence of urethane the respiratory rate was reduced, and the temperature coefficients were lowered. The degree of lowering of the temperature coefficient was related to the degree of suppression of the respiration. In the presence of 3.0 per cent urethane (which at 25°C. diminished the respiratory rate to 42 per cent of normal) the Q_{10} was reduced to 1.3. Lower concentrations of urethane produced Q_{10}'s intermediate between that value and the control value of 2.5.

Thus, temperature coefficients of respiration in the Arbacia egg have been experimentally altered through a range corresponding to that between $Q_{10} = 4.1$ and $Q_{10} = 1.3$. Whether still further suppression of dehydrogenase activity by slightly higher concentrations of ethyl urethane will render respiratory rate completely independent of temperature ($Q_{10} = 1.0$) awaits further investigation.

The relation between cellular metabolism and physiological activity.[1]

IRVIN M. KORR. *Department of Physiology, New York University College of Medicine, New York City.*

This is the first progress report of an investigation of the changes in cellular metabolism which take place as certain mammalian tissues pass abruptly from "rest" into physiological activity, e.g., secretion, contraction. We hope that it will ultimately yield considerable information concerning *a*, the functional integration of oxidative mechanisms in the intact cell; *b*, mechanisms of energy transfer, and *c*, their relation to cellular organization.

Manometric determinations of oxygen uptake have been made on tissue slices, before and after stimulation by specific humoral agents (e.g., salivary glands stimulated by acetylcholine and adrenalin, pancreas by secretin, myometrium by oxytocin), in the presence of specific enzyme inhibitors, activators and substrates. In this manner the degree of participation of certain key enzymes or enzyme systems can be delineated for the resting and for the functioning tissues.

Such experiments with azide and cyanide, together with direct spectroscopic observation of cytochrome bands, have indicated that little or none of the respiration of the resting cell proceeds through the cytochrome-oxidase system. Upon stimulation of the tissues large increases in respiration take place which are mediated by the Warburg-Keilin system, and are prevented or eliminated by the above poisons.

The manner in which the Warburg-Keilin system becomes engaged on stimulation, and other oxidative changes are now under investigation in the submaxillary gland.

Experiments with paraphenylenediamine indicate that the Warburg-Keilin system is present in the resting cells in a form capable of reaction, but that it depends, for its participation in the respiratory complex, upon an agent or agents capable of reducing the cytochrome, and that this

[1] Aided in part by grants from the American Philosophical Society and from the Ella Sachs Plotz Foundation.

agent is made available upon stimulation of the cells; it can be abruptly withdrawn by reversal of the stimulation (by atropine). This agent must function in connection with the substrate-dehydrogenase systems.

Shifts in the latter also occur. For instance, fluoride does not reduce the respiration of the unstimulated cell, whereas it prevents the normal rise in metabolism (and the "gearing" of the Warburg-Keilin system) when acetylcholine is added.

Closer identification of the systems involved and of their interrelations is now in progress.

Urine dilution and concentration tests in normal and adrenalectomized dogs. F. J. KOTTKE (by invitation), C. F. CODE and E. H. WOOD (by invitation). *Department of Physiology, University of Minnesota, Minneapolis.*

The power of the kidney to elaborate a dilute urine and a concentrated urine was tested in adrenalectomized dogs maintained in good condition, without cortical hormone, on a high sodium-low potassium diet. Under similar conditions the specific ability of the kidney to concentrate chloride has been studied. The performance of the kidney during these tests was compared with that of normal dogs maintained on the same diet.

The specific gravity of the urine was used to measure the diluting and concentrating ability of the kidney. In performing the dilution test 300 cc. of distilled water was given to the fasted dog by stomach tube, and urine samples were obtained at hourly intervals. The concentrating ability was tested by depriving the animal of food and water and determining the specific gravity of the urine during a number of twelve hour periods. To appraise the power of the kidney to dispose of excess chloride, the urine chloride concentration was determined at ten minute intervals over a period of several hours following the intravenous injection of 350 cc. of 4.5 per cent NaCl solution. All urine samples were obtained by catheterization.

If the adrenalectomized animal was well sustained on the high sodium-low potassium diet without cortical hormone, as dilute or nearly as dilute a urine was obtained as from the normal animals. However, even though the adrenalectomized animals were well maintained without hormone the power of their kidneys to concentrate urine was reduced. Similarly, the ability of the kidney to dispose of excess chloride by high urine chloride concentrations was limited by the lack of cortical hormone.

Simultaneous observations on the activity of the gall bladder, the sphincter of Oddi and the duodenum. D. D. KOZOLL[1] (by invitation) and H. NECHELES. *Department of Gastro-Intestinal Research, Michael Reese Hospital, Chicago, Ill.*

This work was undertaken to study the emptying mechanism of the gall bladder. Since this mechanism obviously depends on three main factors, these were investigated in acute experiments on anesthetized dogs. These three factors are: motility and tone of the gall bladder, of the duodenum, and the resistance of the sphincteric mechanism of the duodenal portion of the common duct. Balloons were inserted into gall bladder and lower duodenum and their activity recorded by manometers.

[1] Bernard Portis Research Fellow in Surgery.

The common bile duct was sectioned; its proximal part was intubated for the collection of bile; its distal part was cannulated and connected to a suitable manometer; through a T tube attached to the latter, isotonic saline solution was perfused at a constant rate through the common duct into the duodenum. A cannula permitted the outflow of the saline from the duodenum. The resistance offered to the flow of saline through the sphincter of Oddi was recorded by manometer. Respiration was recorded by a balloon inserted between the diaphragm and liver. Blood pressure was recorded by the usual method.

Results. The sphincter of Oddi has a regular and distinct tonus rhythm, independent of the tonus and of the gall baldder and duodenum. There is a surprising lack of correlation between high tonic contractions of duodenum and the tone of the sphincter of Oddi, i.e., subtetanic contractions of the duodenum frequently do not impede the full flow of perfusion through the sphincter of Oddi. The motility of the gall bladder is independent of the spontaneous activity of duodenum and sphincter of Oddi. The intravenous administration of 200 cc. of 0.9 per cent saline solution is followed consistently by marked increase of sphincter tone and after a short interval by marked increase of duodenal tone and motility. The intravenous administration of 200 cc. of 5 per cent glucose solution never had any effect on the sphincter and usually inhibited the duodenum. Neither saline nor glucose solution had an effect on the gall bladder. The effects of saline glucose mixtures, hypertonic solutions and painful stimuli will also be reported.

The glass spoon manometer for optical pressure recording. WILLIAM G. KUBICEK (by invitation), FREDERICK P. SEDGWICK (by invitation) and MAURICE B. VISSCHER. *Department of Physiology, University of Minnesota, Minneapolis.* (Demonstration.)

The Gibson Spoon Manometer principle has been adapted to physiological research uses. Methods of construction have been described by us. (Review of Scientific Instruments, 1941). Manometers of desired sensitivities over a wide range can be constructed. Instruments of sensitivity suitable for arterial pressure recording have natural frequencies ranging from 100 to 160 cycles per second under operating conditions.

Manometers will be demonstrated having sensitivities ranging from 0.2 to 2.5 mm. deflection per mm. Hg at 2 M distance. The angular deflection is linear with pressure and the calibration does not change over several months of use. Elastic hysteresis is most pronounced with soft glass, less with pyrex and least with quartz. The correction required on this account, even with soft glass, is relatively small.

Experimental coronary insufficiency in the dog: electro-cardiographic, blood pressure and pathologic study. JOSEPH J. LALICH[1], GEORGE W. WALKER, and LOUIS COHEN (introduced by O. O. Stoland). *Hixon Laboratory for Medical Research, University of Kansas, Kansas City.*

Coronary insufficiency was studied in 26 dogs for periods of a few days to one year. Analysis of 292 normal grams on 26 dogs showed that inverted or diphasic (minus plus minus) T waves are most frequent and that RS-T deviation occurs in 4.1 per cent of the records. Sodium pentobarbital

[1] George A. Breon Fellow in Experimental Medicine.

in anesthetic doses causes tachycardia, may produce RS-T deviation, and may make positive T waves inverted. In 18 dogs the left circumflex coronary was dissected and partially constricted with a Goldblatt clamp. In eight control dogs two had their pericardia cut, six had the circumflex artery dissected, and clamps were applied without constriction to three dogs. Some of the control dogs developed positive T waves, and minor RS-T deviation for 1 to 3 days. The constricted dogs in 270 records showed RS-T deviation, loss of Q1, and upright T waves. Arrhythmias appeared in 12 to 24 hours and lasted but two days. Normal grams followed in 5 to 10 days and remained from one to four months when Q1 and T wave changes, with RS-T deviations reappeared and persisted from one to six months. Blood pressure studies (Hamilton manometer) on 464 records revealed that coronary constriction did not alter the arterial blood pressure. Pathologic studies showed two out of eight control dogs had infarcts. In the constricted group five had infarcts, five had minimal fibrosis, and eight had no infarction. It is concluded that manipulating the heart or constriction of the circumflex artery will produce EKG changes without infarction.

A method for the study of skin histamine (with some results of splanchnic nerve stimulation). E. H. LAMBERT and S. R. ROSENTHAL (introduced by G. E. Wakerlin). *Department of Physiology and Department of Pathology, Bacteriology and Public Health, University of Illinois, College of Medicine, Chicago.*

Uniform pieces of skin (one cm. in diameter and comparable in thickness to a Thiersch graft) are placed between anode and cathode chambers for electrodialysis; the raw surface of the skin is separated from the cathode fluid by a thin sheet of cellophane. One-half milliampere current is passed through the skin for three minutes. The cathode fluid (0.2 cc. of 0.9 per cent saline) is removed, heated at 100 degrees C. with 0.05 cc. N HCL for 30 minutes, evaporated to dryness, made up to 0.2 cc. with distilled water and adjusted to pH 7.4 with neutral red indicator. The samples thus obtained are assayed on a strip of guinea pig ileum. Samples not boiled in acid contain a substance which counteracts the action of histamine on the gut.

With this method an increase of free histamine in the skin can be detected after intravenous injection of histamine. Large quantities of histamine appear in the cathode fluid when the skin has been superficially burned.

An attempt was made to determine whether histamine is liberated in the skin of the abdomen following stimulation of the splanchnic nerve. Control samples of skin were taken from the right side of the upper abdomen and lower chest in dogs. Electrodes were applied to the left splanchnic nerve under procain or light ether anaesthesia. Following stimulation of the nerve, samples of skin were taken from the left side. Although incomplete, the results suggest that histamine may be liberated reflexly in the abdominal skin upon stimulation of the splanchnic nerve.

Factors governing blood flow through limb vessels.[1] MILTON LANDOWNE[2] (introduced by L. N. Katz). *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.*

[1] Aided by the A. D. Nast Fund for Cardiac Research.
[2] Emanuel Libman Fellow.

Blood flow was determined in the horizontal human leg-foot by a modification of the Hewlett and Van Zwaluwenburg plethysmographic method. The procedure was critically evaluated prior to these studies.

A. Evidence is presented to support the hypothesis (when blood pressure and cardiac output are unchanged) that maximal blood flow in the resting limb is determined not alone by dilatation of the minute vessels but if dilatation of these vessels is greater than a certain degree, maximal flow is then determined also by the diameter of the larger supplying arteries.

This concept is supported by the following observations: 1. When the flow consequent upon release of arterial occlusion was compared to the duration of occlusion, the maximal flow approached a limiting value as the duration was increased. 2. Maximal flow during such hyperemia was of the same order of magnitude as was that induced by local heat. Combined heat and arterial occlusion were no more, or but slightly more, effective than either procedure alone, although the locus of action was not the same. 3. When occlusive vascular disease affecting predominantly the major vessels was present—as in thromboangiitis obliterans—heat, or arterial occlusion, or both, did not induce significant increases in blood flow. In such cases the "resting" flow was "maximal" as well. It is important therefore to consider the diameter of major vessels as a factor which may limit flow through a vascular area. As a corollary the necessity of critically considering all other segments of an area in evaluating flow through any one of these is emphasized.

B. The "reactive" hyperemia consequent to release of arterial occlusion was investigated.

Maximal flow occurred at once in uncongested limbs, was delayed up to 30 seconds in previously congested limbs. Subsidence of increased flow was related to the duration of prior occlusion, and initial vascular state. In some instances after initial maximal flows a sharp transient decrease occurred, succeeded by a secondary rise, and then gradual subsidence.

Simple apparatus for optical registration of vascular dynamics.[1] MILTON LANDOWNE[2] (introduced by L. N. Katz). *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.* (Demonstration.)

Simply designed and easily constructed equipment which make accurate optical recording available for clinical or animal study is presented.

1. A simple light source for multiple optical recording employs a 6-8 volt 32 c.p. automobile lamp, rectangular mask and a focussing lens. The essential feature is the cylindrical plano- or bi-convex lens of 6 to 14 diopters (a cylindrical test case lens or reading glass). This directs a broad beam of sharply focussed light to reflecting and/or focussing surfaces.

2. The intra-arterial monometer of Hamilton is demonstrated, modified into compact form for recording on a electrocardiograph camera. On a single stand are mounted a reservoir connected to the membrane carriage and calibrating manometer. A special adapter and needle are connected by 2 to 3 feet of flexible lead tubing. The system is sterilized with alcohol, filled with sterile 3% sodium citrate and merthiolate 1:100,000.

3. A sterilizable segment capsule is connected by a T tube to a calibration and supply reservoir of sterile sodium citrate and to a needle. This records local venous pressure with the error and damping introduced by

[1] Aided by the A. D. Nast Fund for Cardiac Research.
[2] Emanuel Libman Fellow.

the use of non-rigid tubing. Antecubital venous pulsations may be recorded by this method.

Excitation and inhibition of the inspiratory center by afferent impulses from the lungs. M. G. LARRABEE and G. C. KNOWLTON. *Department of Physiology and Biophysics, Cornell University Medical College, New York City.*

The response of inspiratory motoneurones to changes in lung volume was measured by recording the action potentials of single efferent fibers in the phrenic nerve.

In agreement with Worzniak and Gesell it was found that inflation of the lungs can increase the impulse frequency of the phrenic discharge and that this effect is abolished by cutting the vagus nerves. Such an increase in the frequency of discharge is elicited only by an inflation considerably in excess of the normal inspiratory volume. Furthermore it lasts only a few tenths of a second and is followed by an inhibition of the discharge. Inhibition is the only effect observed with smaller inflations. We interpret this to mean that the excitation and inhibition are mediated by separate sets of afferent fibers which differ in threshold and rate of adaptation.

The reflex effect of normal inflation of the lungs was studied by comparing the discharge of a single fiber of the phrenic nerve during a normal inspiration with that which occurred immediately after producing a pneumothorax. The pneumothorax was made during expiration by opening cannulae previously inserted through the chest wall. The first inspiratory discharge subsequent to the sudden pneumothorax was prolonged, with the impulse frequency increasing along a smooth curve for a time considerably longer than the normal inspiration. The discharge during normal inspiration followed exactly the same frequency curve to a point at which the discharge suddenly slowed, to stop completely after two or three additional impulses.

We conclude that the afferent vagal discharge which develops during eupneic inflation of the lungs exerts no excitant action on the phrenic motoneurones. Neither is there any evidence of an inhibitory action until the discharge suddenly stops.

Effect of previous environmental temperature on the metabolism of the rabbit measured at 28°C. ROBERT C. LEE (introduced by Thorne M. Carpenter). *Nutrition Laboratory, Carnegie Institution of Washington, Boston, Mass.*

The effect of the previous environmental temperature to which rabbits have become acclimated on the metabolism at 28°C. was studied with 12 adult rabbits (1.9 to 5.3 kgm.) in observations over periods of 8 to 18 months. All the metabolism measurements used were made at 28° to 29°C., with the rabbits in complete repose, after they had been 24 hours at 28°C. The metabolism of 5 rabbits, previously living at 17° for 3 weeks, gradually decreased (total decrease 14 per cent) as the stay at 28° increased from 1 to 14 days, but from the 14th to the 30th day it decreased only an additional 3 per cent. When the rabbits were kept 3 weeks longer at 28°, a decrease in metabolism of 2 per cent resulted. When the previous temperature was again lowered to 18° and maintained at this level for 3 weeks, the metabolism measured at 28° increased but was appreciably

lower than that measured after the previous stay at 17°, due to the long adjustment to 28°C. A repeated adaptation to 28° for 3 weeks resulted in an approximate reproduction of the previous metabolic level noted under about the same conditions. A maximum variation of 22 per cent was found between the measurements after the rabbits had been living at 31° and after they had been living at 17°C. With another group of 7 rabbits, the measurements at 28° were 13 per cent lower after the animals had lived at 29° than after they had lived at 10°C. In 89 per cent of the 80 instances where the previous temperature was changed from one level to another, the metabolism at 28° varied inversely with the change in temperature. In exact metabolism studies on rabbits the temperature at which the rabbits live must be carefully controlled. The major adjustment of the metabolic level to the previous environmental temperature takes place within a 3-week stay at the given temperature level. The effect of previous environmental temperature on the metabolism offers a partial explanation of the variability in basal metabolism measurements on the rabbit obtained by different investigators.

Obesity in the rat. PHYLLIS LEVENS (by invitation) and H. G. SWANN. *Department of Physiology, University of Chicago, Chicago, Ill.* (Demonstration.)

Obesity has several times been reported to follow experimental hyperinsulinism in the rat. However, the reported weight gains have not been large. Also, in our hands, a mortality approaching 100 per cent has been found to follow administration to the rat of 6 or more units per day of Protamine Zinc Insulin, if special measures to avoid it are not taken. We have found that if a pellet of Desoxycorticosterone acetate (courtesy of Schering Corp.) is implanted and if 25 per cent cane sugar is made the sole source of drinking fluid, the lethal effects of insulin are counterbalanced and a rapid extreme obesity results. Ten units per day of Protamine Zinc Insulin yield optimum results. Adult rats of 300 grams gain but little further weight under our conditions if untreated. But with this treatment, they gain about 3 grams per day, weights of 600 or more grams being attained. At autopsy, large pads of fat are found subcutaneously and retroperitoneally. Total saponifiable fat of the whole cadaver is increased 3 to 5 fold over the normal.

Some of these obese rats will be demonstrated.

On the non-existence of mitogenetic radiation.[1] JULES LEVINE (by invitation) and ARTHUR H. STEINHAUS. *George Williams College, Chicago, Ill.*

These studies were designed to determine the existence or non-existence of mitogenetic radiation. The original Gurwitsch strain of yeast was used as both detector and sender. The Gurwitsch key experiments on yeast were repeated with negative results.

New methods of detecting possible biological radiations were worked out. Petri dishes of beer wort agar were seeded with a two-day-old yeast culture. Five plates were selected for "radiation" and five for control. At hourly

[1] The conduct of this study was aided by a grant from the American Association for the Advancement of Science and for the period from April, 1936 to August, 1937 by the Works Progress Administration under its project Number 3394.

intervals plugs one centimeter in diameter were punched out from each plate and fixed in 2 cc. of 20 per cent sulfuric acid. Mother and bud cells per plug were counted in a hemocytometer and fourteen-hour growth curves were thus obtained for both "radiated" and control plates. The detector culture was always in the lag phase of growth when exposed to the sender culture. Exposure time, temperature of incubation and distances between sender and detector culture were varied. A series of experiments were done in which yeast was self "radiated" by being exposed to a chromium plate reflecting surface.

Detection of Gurwitsch rays was also attempted by use of liquid yeast capsule and liquid yeast plunger methods. In the capsule method a small quartz capsule containing yeast used as a sender culture is immersed in a test tube containing a detector yeast culture. After exposure the amount of growth in the detector is measured by reading the degree of density in foot candles, as determined by the amount of light which penetrated to a Photoelectric cell. In the plunger method a glass plunger containing yeast sender on honey agar media is inserted in the tube containing the liquid detector culture. After ten minutes exposure the plunger is withdrawn and readings on the exposed culture made every hour.

Analysis of the data obtained from about 350 complete sixteen-hour tests each involving "radiated" and control cultures points to the nonexistence of Gurwitsch or M rays.

The effect of excessive dietary sodium chloride and potassium chloride on the carbohydrate metabolism of normal rats. ROBERT C. LEWIS, JR. and BERNARD B. LONGWELL (introduced by Maurice H. Rees). *Department of Biochemistry, University of Colorado School of Medicine, Denver.*

The utilization of administered carbohydrate has been compared in male albino rats maintained during a two week preliminary period on diets of varying sodium chloride and potassium chloride content. The concentrations of *added* salts in grams per kilo were as follows: control diet, 0.69 grams of sodium, 2.38 grams of chlorine and 2.99 grams of potassium; high sodium chloride diet, 33.7 grams of sodium, 49.7 grams of chlorine and no *added* potassium; high potassium chloride diet, no *added* sodium, 22.5 grams of chlorine and 26.5 grams of potassium.

The fasting blood sugar levels showed about the same range in the rats on each of the three diets. However, the tolerance to orally administered glucose and the sensitivity to subcutaneously administered insulin was greater in the high sodium chloride animals than in the controls. Although the glucose tolerance of the high potassium chloride animals did not differ significantly from that of the controls, the sensitivity of these animals to insulin was decreased. Insulin sensitivity was judged by the initial rate of fall of the blood sugar, by the incidence of insulin reactions and by the rate of return of the blood sugar toward the fasting level.

The observed differences in glucose tolerance may have been due to an alteration either in the rate of oxidation of glucose or in the rate of its storage as glycogen. The animals on the high sodium chloride diet oxidized less of an administered dose of glucose than did the controls, and the animals on the high potassium chloride showed no significant variation from the controls in this respect. Thus, there was presumptive evidence

that the observed changes in glucose tolerance and in insulin sensitivity were brought about by storage changes rather than by alterations in the oxidative reactions. However, although part of the evidence obtained by direct measurement of glycogen storage in the muscles and livers confirmed this view, the results in this phase of the study were not entirely consistent.

Papaverine and ventricular fibrillation.[1] E. LINDNER (by invitation) and L. N. KATZ. *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.*

In a series of five dogs in which the coronary circulation was maintained by perfusion with defibrinated dog blood, papaverine hydrochloride (Lilly) in concentrations of 1:200 to 1:340 in the coronary vessels resulted in the disappearance of faradically induced ventricular fibrillation and the restoration of synergic beating within five minutes of its administration. In three other dogs without such coronary perfusion, the intraventricular administration of papaverine hydrochloride accompanied by manual massage changed ventricular fibrillation to synergic beating within 22 minutes. Massage without papaverine in two other dogs failed to abolish the fibrillation.

In a series of 8 dogs, production of ventricular fibrillation by faradic stimulation required definitely greater current strength after administration of papaverine than before. The strength of stimulus required for ventricular fibrillation in the untreated dog was that obtained from a Harvard inductorium when the secondary coil was 10 cm. from the primary. After the administration of papaverine, the stimulus required for fibrillation was that obtained when the secondary coil was only five cm. from the primary. After reestablishment of synergic beating in the animals treated with papaverine, the current strength required for refibrillation was greater than in the controls.

The influence of pyramidal excitation on the spinal cord of the cat. DAVID P. C. LLOYD. *Laboratories of The Rockefeller Institute for Medical Research, New York City.*

Stimulating electrodes were placed on the medullary pyramids between a transection cranially and a lesion excluding the pyramids caudally. Thus cortical reverberation and extrapyramidal activity were excluded. Records were obtained by microelectrodes in the cord, or by ventral root leads.

Single shocks cause: 1, a tract response which in the lumbar cord begins about 4.5 msec. after the shock and lasts several milliseconds, representing conduction velocities downward from 65 M./sec., and 2, a burst of internuncial activity, consisting of small spikes (50 μV), which is most prominent in the extreme lateral intermediate region, and lasts 40 msec. Tetanic stimulation intensifies this internuncial activity. Subsequently large spikes (1–2 mV.) appear after 10–20 msec. at the base of the dorsal horn and in the ventral intermediate region. Similar dorsal horn spikes are initiated by dorsal root volleys, and interaction between pyramidal and primary afferent activity is demonstrable at this point. The ventral

[1] Aided by the A. D. Nast Fund for Cardiac Research.

intermediate spike activity is paralleled by the course of facilitation in the motoneuron pool.

Since little or no motoneuron discharge accompanies short duration tetani, facilitation of reflex discharges is used to detect pyramidal influence on motoneurons. Facilitation begins 9–10 msec. following the onset of the pyramidal tetanus. At first facilitation is confined to arcs of three or more neurons. Only after 12–20 msec. is the two-neuron arc facilitated. It would seem, therefore, that motoneurons are influenced secondarily to activation of the intermediate region. Indirect action of the pyramidal system on motoneurons is confirmed for the more rapidly conducting pyramidal fibers by comparing motoneuron facilitation curves obtained with successively longer trains of pyramidal shocks. The added effect of each successive volley is exerted only after 5.5–6.5 msec. latency. This represents a considerable reduction of the summation time from the first shock to the first detectible facilitation, but still necessitates the inclusion of at least one interneuron in the pathway. It is possible that the more slowly conducting pyramidal fibers affect motoneurons directly, but the resultant must be small by comparison, since no measurable facilitation occurs following single or paired pyramidal shocks.

Hepatic dysfunction in relation to the reaction between blood serum and colloidal gold. EARL R. LOEW and PAUL NOTH (by invitation). *Wayne University College of Medicine, and Detroit Receiving Hospital, Detroit, Mich.*

It has been indicated (Gray, S. Arch. Int. Med. **65**: 524, 1940) that the presence of liver disease is demonstrated when blood serum flocculates colloidal gold under specified conditions. The sensitivity and specificity of the colloidal gold liver function test suggests great usefulness in the clinical and experimental laboratory.

With the colloidal gold technique we have tested sera from 25 medical students and faculty members and 191 patients at Detroit Receiving Hospital. This report includes data from all patients studied except those known to be receiving chemotherapy. Patients were examined by the clinical author (P. N.) and the clinical diagnoses were verified whenever possible by biopsy material or at operation or autopsy.

Positive gold reactions were obtained in 87 per cent of 45 patients of which 23 were diagnosed as hepatic cirrhosis and 22 diagnosed as acute or subacute hepatitis. Not more than 75 per cent of the bromsulphalein retention or hippuric acid tests were positive in patients giving positive gold reactions. The incidence of positive gold reactions was 60 per cent in 10 cases of primary or secondary hepatic carcinoma, and 33 per cent in 27 patients with congestive heart failure with and without definite evidence of secondary liver involvement. Only 2 positive gold reactions were encountered in 19 cases of acute or chronic cholecystitis with or without cholelithiasis. Positive gold reactions occurred in approximately 75 per cent of patients with miscellaneous liver diseases or those with liver involvement, the nature of which was not definitely diagnosed.

In a control group of 68 hospitalized patients with no laboratory or definite clinical evidence of hepatic disease the incidence of positive gold reactions was 9.4 per cent. No positive gold reactions were obtained with sera from 25 normal individuals. The high incidence of positive gold

reactions in certain types of liver disease suggests the use of the gold reaction as a routine diagnostic aid.

Comparative studies of the respiratory act (activity patterns). G. N. LOOFBOURROW (by invitation) and ROBERT GESELL. *Department of Physiology, University of Michigan, Ann Arbor.*

Breathing in the chicken differs importantly from that in mammals. In the dog most, or even all, of the energy for both inspiration and expiration of air may be provided by the inspiratory muscles. In the chicken, the work of inspiration and expiration seems relatively evenly divided between the opposing groups of muscles. This difference is related to anatomical arrangements of the respiratory tracts. In the dog, all of the air is inspired into the lungs. In the chicken, much additional air is inspired through the lungs into the flaccid thoracic and abdominal air sacs. In the mammal, potential energy is stored by distortion of the lungs and torso sufficient to expel the inspired air during relaxation of the inspiratory muscles. In the fowl, active expiratory contractions empty the air sacs through the lungs during the phase of expiration. In the dog the opposing groups of muscles show individualistic patterns of activity. The inspiratory muscles exhibit the slowly augmenting and rapidly weakening pattern of activity while the expiratory muscles show either the steady state or rapidly augmenting and slowly weakening pattern. In the chicken both inspiratory and expiratory contractions show the slowly augmenting inspiratory pattern of activity characteristic for the inspiratory muscles of the dog, mouse, rat, rabbit, and horse. Frequency of muscle fiber twitch and recruitment of newly activated units were found to increase with the progress of each act. It is, therefore, concluded that the mechanism of gradation of the respiratory contractions of the chicken necessary to meet the mechanical requirements of the respiratory act is the same in both inspiratory and expiratory muscles as that previously described in detail in the inspiratory muscles of dog.[1]

Preliminary experiments on the alligator indicate that the inspiratory pattern of activity of the diaphragm is of the slowly augmenting type found in the mammals.

The action of angiotonin on the completely isolated mammalian heart. VICTOR LORBER (by invitation) and MAURICE B. VISSCHER. *Department of Physiology, University of Minnesota, Minneapolis.*

The action of angiotonin was studied on the cat heart, completely isolated according to the method described by Moe and Visscher (Am. J. Physiol. **125:** 461, 1939). Nine experiments were done, in six of which oxygen utilization was measured continuously in a closed system. In all the experiments cardiac output, coronary flow, and changes in diastolic volume were measured. Filling and emptying pressures of the heart were controlled, but diastolic volume was permitted to vary.

It was found that under the circumstances of these experiments, angiotonin markedly constricted the coronary vessels for very short periods, an observation already reported by others (Hill and Andrus. Proc. Soc. Exper. Biol. and Med. **44:** 213, 1940). In addition, however, it caused

[1] Gesell, R., A. K. Atkinson and R. C. Brown. Am. J. Physiol. **131:** 659, 1941.

regularly a prolonged decrease in diastolic volume, as well as a marked and extended increase in work performance and efficiency.

Although there was no exact parallelism between the effects on the coronary vessels and myocardium at larger doses, both effects decreased in intensity and disappeared as the dosage was diminished. In two cases in which one effect was elicited independently of the other, it was the coronary effect that failed to appear. The myocardial action in one instance made itself evident in a small, transient decrease in diastolic volume, and in the other, through a small, though protracted increase in work performance and efficiency, as well as a decline in volume. In both cases it is possible that a small and transient coronary flow effect may have occurred between measurements.

It seems likely, on the basis of these experiments, that if angiotonin is formed in vivo in sufficient quantities to produce a vascular effect, it will probably also exert its stimulating effect on the myocardium.

We are indebted to Dr. Irvine H. Page for suggesting these studies and for furnishing the angiotonin solutions.

Electrotonus produced by direct current pulses in frog nerve. R. Lorente de Nó and L. Davis, Jr. (by invitation). *Laboratories of The Rockefeller Institute for Medical Research, New York City.*

In freshly excised nerve the anelectrotonus and the catelectrotonus are symmetrical. They are quite satisfactorily described by Hermann's model with fixed resistance and capacity (CR of the order of one msec.).

After standing for some time in Ringer's solution, or for a few seconds in 5 per cent CO_2, another slow component of the electrotonus becomes prominent. Its rising phase at a few millimeters from the electrodes may last from several tenths of a second to one second or more; often after the rising phase the membrane potential drops to a steady lower level. When this happens, the membrane potential, after the end of the pulse, reverses its sign; decremental oscillations may then follow. In one case oscillations of about five seconds' period, lasting for half a minute, were observed.

Mathematical treatment of a variety of possible physical models of nerve indicates that nerve acting as a passive structure could not exhibit a behavior such as has been described. Therefore, during the slow component of the electrotonus the membrane must add to the polarizing potential its own reaction, consisting of a change of its e.m.f. in the opposite direction of the polarization. Apparently the nerve expends energy in order to remain near its equilibrium state.

The slow electrotonus spreads farther than the fast one. The anelectrotonus is usually, but not always, stronger than the catelectrotonus. Conduction of a train of impulses or ether anesthesia reversibly abolishes the slow component, leaving the symmetrical unchangeable fast component.

The slow component is intimately related to the after-potentials and to the residual potentials that appear at the margin of and beyond a conduction block.

The crystallization of cholesterol from bile and its relation to the formation of human gallstones. Marie Lorenz (by invitation) and K. K. Jones. *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.* (Demonstration.)

The crystallization of cholesterol from fatty acids and bile residues, the reduction of biliverdin to bilirubin by fatty acids, and the relation of this to gall stone formation, will be demonstrated.

The human skin as a conductor of 60 cycle alternating current of high intensity, studied on "electroshock" patients. HANS LÖWENBACH and JASPER E. MORGAN (introduced by G. S. Eadie). *Department of Neuropsychiatry and Physiology, Duke University School of Medicine, Durham, N. C.*

The treatment of mental disorders with electric "shock" provided a new opportunity to examine the living human skin as a conductor of a 60 cycle alternating current of relatively high intensity (300–1000 milliamperes). In a first series of experiments (31 individuals with 98 single "shocks") an ink-writing oscillograph recorded the impedance offered to a potential of about 20 microvolts. The electrodes (round copper plates, 5 cm. in diameter, electrode jelly between metal and skin) lay on both temples. A second ink-writer served as a recording milliammeter of the "shock" current which was applied through the same electrodes. In a second series the "shock" current was recorded with an oscillograph able to follow frequencies up to 15000 cycles for detailed analysis of amplitude and waveform. Finally, human corpses shortly after death, and excised skin for a period of several weeks were tested for their conductance.

When 50–120 volts are given for 0.1–0.4 sec., the impedance drops instantaneously to a value between 270 and 120 ohms regardless of the original impedance, if the latter was kept at or below 2000 ohms. In females the decrease is slightly smaller than in males. During the time of current-flow there is no further gross change of the impedance. The waves do not deviate perceptibly from the sine form. When the current is broken, the impedance—to 20 microvolts—rises suddenly by 80–150 ohms, depending on the "shock"-tension applied; it then increases over a period of many seconds, approaching the original value asymptotically. The return to pre-"shock" impedance takes place faster after low tensions than after high ones. A second "shock" given while the impedance is still low, produces the same current as the previous one.

The skin of human corpses reacts similarly to that of living individuals. However, the impedance drop is less with increasing interval between the time of death and of examination. Excised skin gradually loses the properties of the living skin and, finally, conducts like other organic material, i.e., it follows predictably the laws of electrolytic conduction.

Phosphorus metabolism of the musculature of E-deficient suckling rats. GWEI DJEN LU (by invitation), GLADYS A. EMERSON (by invitation) and HERBERT M. EVANS. *Institute of Experimental Biology, University of California, Berkeley.*

Inorganic phosphate, creatine phosphate and total acid soluble phosphate were determined in the entire musculature of the hind limbs of 33 suckling rats from vitamin E-deficient mothers. They were sacrificed when from 22 to 31 days old; in all cases the young were paralyzed. As controls, similar studies were made of the musculature of 8 sucklings from normal mothers and of 4 suckling from mothers receiving 15 mgm. of alpha-tocopherol on the day of littering. Six young were studied two days after spontaneous recovery from severe paralysis had occurred.

In the paralyzed rats there was a slight decrease in inorganic phosphorus and in creatine phosphorus, and a marked decrease in total acid soluble phosphorus and pyro and organic esters phosphorus, the last mentioned decrease accounting for about 65 percent of the decrease in total acid soluble phosphorus. It is to be noted, however, that the ratio of the inorganic and creatine phosphate phosphorus to the total acid soluble phosphorus was not altered in spite of a decrease in muscle creatine. It is to be noted that in the paralyzed cases, not markedly dystrophic, no change occurred in the calcium content. The ratio of creatine to phosphorus lost is not equamolar. More phosphorus is allowed to circulate in inorganic form.

In the case of two markedly dystrophic sucklings, different changes were found in the musculature. The limb muscles of these animals were white and waxy in appearance and microscopically profoundly degenerated. There was a 50 to 180 percent increase in calcium and great increase in total acid-soluble phosphorus. These changes were accompanied by a greater decrease in phospho-creatine phosphorus.

The ability to phosphorylate glycogen was decreased about 40 percent in the acutely paralyzed group and over 80 percent in the markedly dystrophic group. In vitro additions of vitamin E did not restore the esterification of glycogen.

Pituitary-diabetes in the cat: recovery under phlorhizin treatment. F. D. W. Lukens and F. C. Dohan (by invitation). *George S. Cox Medical Research Institute, University of Pennsylvania, Philadelphia.*

Following the announcement that pituitary-diabetes could be produced in the normal dog (F. G. Young, 1937) attempts were made to extend this method to the cat. We, like Young, have failed to do this in normal cats. However, after removal of part of the pancreas, leaving enough to prevent spontaneous diabetes, it has been possible to make such animals permanently diabetic by a short course of anterior pituitary extract. A preliminary report has been made (Science **92:** 222, 1940) on the course of the island lesions in this species and on the morphological and physiological recovery of the islands when insulin treatment was initiated in the first 3 months of diabetes. The response to other procedures was studied in order to determine what particular aspect of insulin action was responsible for the reversal of the island damage. Phlorhizin, which causes glycosuria in the presence of a low blood sugar, was first investigated. F. M. Allen (1923) had shown that this drug could lower the blood sugar of partially depancreatized diabetic animals. It has been found that phlorhizin (0.2 gram per day) would lower the blood sugar of pituitary-diabetic cats. On a constant diet, the glycosuria during phlorhizin administration was but little increased above that of the preceding pituitary-diabetes. Phlorhizin was given for two weeks. When it was discontinued the urine became sugar free in 5 to 10 days and the blood sugar remained normal. Morphological recovery of the islands has been observed along with this functional recovery from diabetes. Although the details of phlorhizin action require further study the relation of the low blood sugar level to the recovery of the islands under both insulin and phlorhizin is noteworthy. This is a preliminary report.

Factors in the revival of Anguillula aceti after its solidification in liquid air. B. J. Luyet and M. C. Hartung (by invitation). *Department of Biology, St. Louis University, St. Louis, Mo.*

It has been reported previously that the vinegar eel, *Anguillula aceti*, a nematode about 2 mm. long, can be revived after having been immersed in liquid air (−195°C.) if its water content has been partially reduced by immersion in 30 per cent ethylene glycol and if the worm is rewarmed at a velocity of the order of 1000 degrees per second. The reason of the necessity of so rapid a rewarming is that an exposure of even a tenth of a second to temperatures immediately below zero allows the formation of ice and is fatal. The present report is concerned with the extent of this zone of dangerous temperatures. The worms, after immersion in liquid air, were exposed for 5 minutes in an iso-pentane bath maintained at a constant temperature from −50° to −25°C., then they were rapidly rewarmed. None of the animals exposed to temperatures above −40° survived, while, out of 80 exposed to temperatures below −43°, 28 survived. The dangerous zone (that of the formation of ice) for the tissues of *Anguillula aceti* must, therefore, extend from 0° to about −40°C.; at lower temperatures these tissues can be kept in the vitreous state, at least for 5 minutes.

Action of snake venoms on isolated bronchi. DAVID I. MACHT. *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md.* (Read by title.)

Solutions of various snake venoms, prepared by dissolving the dried scales in physiological saline, were studied on bronchial muscle strips from pigs and bronchial rings from cats and rabbits and also on lung preparations from frogs. Cobra venom solution (H. W. & D.), consisting almost entirely of neurotoxin and practically free from hemotoxic and proteolytic constituents, had no effect on either the tonus or contractions of the bronchial muscle preparations. Treatment of these preparations with cobra neurotoxin did not interfere with subsequent action on the same rings or strips of such well-known pharmacodynamic agents as epinephrine, pilocarpine, physostigmine, atropine, papaverin and barium chloride. A solution of *crude* cobra venom containing all the original constituents either did not affect or but slightly relaxed the tonus and also did not influence the response of such preparations to the other pharmacodynamic reagents. The effects of viper venoms differed from those of cobra venom. Five to 10 mg. of scales of *Crotalus* venom and of the venom of Russell's viper dissolved in 50 cc. of Locke solution markedly relaxed the preparation but did not later inhibit response of these bronchi to treatment with the pharmacodynamic reagents mentioned above. The difference between the venoms of the vipers and that of the cobra may be explained in part at least by the relatively smaller content of the latter in respect to cytotoxic and proteolytic constituents.

Effect of cobra venom on isolated uterus. DAVID I. MACHT. *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md.* (Read by title.)

The effects of both crude cobra venom and cobra neurotoxin were studied by the usual physiological methods on the tonus and contractions of isolated uterine strips from virgin and pregnant guinea pigs, rabbits and cats in oxygenated Locke solution. Solutions of cobra neurotoxin (H. W. & D.) in doses of from 5 to 50 mouse units in 100 cc. of Locke solution diminished neither the tonus nor the contractions of the preparations; on the contrary, in some cases the tonicity of the uterine muscle was definitely increased. Crude venom containing hemotoxic and proteolytic principles exerted the

same effect. In no instance was failure of the muscle to respond to such powerful pharmacodynamic agents as epinephrine, pilocarpine and atropine observed after its exposure to the venom for from 10 to 15 minutes. This lack of interference of cobra venom solution with the contractions of uterine muscle previously exposed thereto is contrasted with the effect that morphine exerts on such a preparation. Clinical reports from several obstetricians who have employed cobra venom as an analgesic in labor confirm the writer's laboratory findings. When administered to patients, cobra venom produced some analgesia without inhibiting contractions of the uterus.

Localization of cobra venom analgesia. DAVID I. MACHT. *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md.* (Read by title.)

That relief of pain effected by cobra neurotoxin which makes it a useful substitute for morphine in advanced malignant disease and other forms of intractable pain reveals such similarity and dissimilarity between morphine and cobra venom as aid materially in localizing the venom's point of action. Cobra venom does not anesthetize peripheral sensory nerve terminals locally nor in ordinary therapeutic doses does it block conduction of ascending or descending nerve fibers. It exerts neither synergism nor antagonism in combination with such convulsants as strychnine, which act on the spinal cord. The analgesia produced thereby is therefore of central cerebral origin. Unlike morphine, however, the cobra neurotoxin reduces neither the acuity nor the field of vision and does not depress the auditory and olfactory senses. Experimental psychological studies on human individuals proved conclusively that ordinary doses of the cobra neurotoxin do not have a narcotic effect on the cerebral cortex or interfere with intellectual performance; on the contrary, they usually stimulate mental efficiency like caffeine. Other psychological and pharmacological experiments such as those on the action of the drug on temperature regulation, on pain thresholds measured quantitatively in rats, guinea pigs, and normal human beings, on the behavior of rats in a circular maze, on reactions of human subjects to tests for neuromuscular responses and coordination, on psychogalvanic reflexes and on basal metabolism all seem to indicate that the analgesia produced by cobra venom is largely localized in the hypothalamic region and that this depressant effect does not spread to the cortex and particularly to the frontal lobes of the cerebrum as does that produced by the opium narcotics. These experimental laboratory findings on lower animals and normal human subjects are corroborated by numerous controlled clinical reports from physicians who relieved pain in cases of cancer, neuralgia, etc., by injecting cobra venom without stupefying either the special senses or the acumen of the patients.

The effect of high frequency mechanical vibrations on cocaine and cobra venom. DAVID I. MACHT and GUSTAV BERGSON (by invitation). *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md., and Cruft Physical Laboratory, Harvard University, Cambridge, Mass.* (Read by title.)

Prompted by Macht's findings with regard to the influence of radiations on drugs (Am. J. M. Sc. **109:** 520, 1935; J. Am. Chem. Soc. **49:** 2017,

1927; Arch. f. exper. Path. u. Pharmakol. **146**: 177, 1929), the present writers initiated an inquiry into the effects on drugs of exposure to mechanical excitation of supersonic frequencies. Two diverse types of drugs were selected: 1, cocaine, an alkaloid the chemical structure and cleavage products of which are well known, and 2, cobra neurotoxin, a new analgesic, the chemistry of which is yet unsolved and which is more stable than cocaine at ordinary temperatures. Solutions of physiologically assayed cobra venom in saline and of cocaine hydrochloride in distilled water, sealed in hard-glass ampules, were exposed to mechanical vibrations of high frequencies for various times between 10 and 180 minutes. The physical setup was a Pierce generator consisting essentially of a vacuum tube oscillator circuit generating the desired frequencies and a vacuum tube amplifier. The effects of a wide scale of frequencies, ranging from 18 to 70 kilocycles per second, were studied. Changes in cobra venom were measured by bioassay on mice. The effects on cocaine were tested for toxicity on *Lupinus albus* seedlings (J. Gen. Physiol. **4**: 573, 1922) and on mice, and for local anesthesia on rabbits. Certain frequencies of excitation over periods of 40 to 180 minutes produced definite changes in physiological potency of both drugs. Particularly effective were treatments with frequencies of 36 and 48 kilocycles per second. Exposure of cobra venom to 48 kilocycles for 40 minutes reduced its potency for mice from 10 to 5.5 mouse units per cubic centimeter. A 0.5 per cent solution of cocaine hydrochloride, excited for 180 minutes at 36 kilocycles, gave a phytotoxic index of 64 per cent, as compared with 104 per cent, derived from the controls, when tested on growth of seedlings in 1:5,000 solutions, and lost most of its local anesthetic action. A comparison of the results obtained by high frequency excitations with the effects of thermal agitation is in progress.

Influence of estrone and progesterone on seedlings of Lupinus albus. DAVID I. MACHT and DOROTHY J. BROOKS (by invitation). *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md.* (Read by title.)

Crystalline estrone, estradiol and progesterone were first dissolved in 90 per cent ethyl alcohol. From this alcoholic solution concentrations of the crystalline hormones ranging from 1:100,000 to 1:5,000,000 were made in plant-physiological saline solutions (Shive) and growth of *Lupinus albus* seedlings therein for periods of twenty-four hours at 15°C. in the dark was carefully studied by measuring the increment of their roots in the manner described elsewhere by Macht (J. Gen. Physiol. **4**: 573, 1922). Estrone and estradiol, in concentrations of from 1:1,000,000 to 1:5,000,000, stimulated root growth. These results obtained with hydroponic cultures of seedlings agree with horticultural experiments reported by Liebe and Neurath (Biochem. Ztschr. **289**: 198, 201, 1936–37). Progesterone in the same concentrations definitely *inhibited* root growth, the phytotoxic index of *Lupinus albus* seedlings grown in such solutions ranging from 60 to 80 per cent. Mixtures of estrone and progesterone generally resulted in synergistic phenomena, the combination of the two producing a more toxic index of growth than could be accounted for by simply adding the effects of the individual components.

Physiological and toxicological effects of some fish muscle extracts. DAVID I. MACHT, DOROTHY J. BROOKS (by invitation), and ELIZABETH C. SPENCER (by invitation). *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md.* (Read by title.)

The possibility of elaboration by fishes of poisonous substances not of putrefactive origin has been explored to but a limited extent. Mosso found that the blood serum of eels was poisonous and Japanese investigators have extracted a toxin known as Fugu poison from the ovaries and testes of certain species of *Tetraodontidae* and *Diodontidae*. The present writers studied the physiological effects of *muscle juices* of a large series of both edible and inedible fishes. Fresh muscle was ground with equal parts by weight of sand and physiological saline in the proportion of 2 cc. for each gram of tissue. This emulsion, filtered through fine linen, was studied by zoöpharmacological methods on mice and phytopharmacological tests on *Lupinus albus* seedlings. Of the seventy varieties of fish examined, the majority were not poisonous but a number of them yielded extracts that were toxic for both mice and seedlings. Such poisonous muscle extracts were obtained from the catfish, eel, skate, stingaree, toadfish, balloon fish, porcupine fish, bur-fish and moonfish (*Selene vomer*). Two cubic centimeters of these extracts, injected intraperitoneally or intravenously, poisoned and usually proved fatal for mice; and one per cent solutions in plant-physiological saline gave phytotoxic indices ranging from 30 to 60 per cent, while most of the edible fish muscle extracts yielded from 80 to 110 per cent. The possible relationship of these ichthyotoxins to other animal poisons, particularly snake venoms, is now being studied. Most of the toxic muscle extracts were obtained from scaleless fishes, thus giving scientific support to the ancient classification of edible and inedible fishes into those which have scales and those which have none.

Muscle extracts from some "shellfish" (not true fish but either mollusks or crustaceans) were also examined. Muscle juices from squids, lobsters, shrimp, scallops, clams and oysters all proved toxic for mice on intraperitoneal injection and gave phytotoxic indices for *Lupinus albus* ranging from 30 to 60 per cent.

Phytotoxic reactions of blood sera from psychotic patients. DAVID I. MACHT, MOSES B. MACHT (by invitation), and LESTER L. BURTNICK (by invitation). *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, and Springfield State Hospital, Sykesville, Md.* (Read by title.)

A systematic study, by Macht's special phytopharmacological methods (described in previous publications—"Protoplasma", xxvii, 1, 1937; J. Lab. and Clin. Med. **26:** 597, 1941), of fresh blood sera obtained from patients suffering from various types of mental disease has been initiated by the present authors and is still in progress. They have found that from 80 to 90 per cent of the sera thus obtained are definitely toxic for roots of *Lupinus albus* seedlings grown in one per cent solutions thereof in plant-physiological saline at 15°C. in the dark, as compared with numerous controls grown in physiological saline and in normal human blood sera. A statistical analysis of the findings shows that the figure expressing the difference in toxicity between normal and psychotic blood sera is not only very great but absolutely reliable. Psychotic sera were obtained from a

large variety of psychotic patients, including manic-depressive, schizophrenic, melancholic, paretic and other groups. One hundred cases have so far been studied. The most striking finding made in this investigation up to the present time is that phytotoxic reactions are effected by sera from both the so-called "organic" and "functional" psychotic cases so that in that respect there is no sharp line of demarcation between the two groups.

The influence of cobra venom and opium alkaloids on behavior of fish. DAVID I. MACHT and ELIZABETH C. SPENCER (by invitation). *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md.* (Read by title.)

Inasmuch as the analgesia and the effect of cobra venom on the behavior of rats in the maze do not run parallel in contrast to the action of the opium alkaloids on the behavior of rats (M. B. Macht. Proc. Soc. Exper. Biol. and Med. **42:** 436, 1939), the writers determined to study the effects of these drugs also on fish. Kymographic records or ichthyograms traced by goldfish, *Carassius auratus* (two inches long), placed in solutions of cobra venom and various opium alkaloids, respectively, were made by a technique which the senior author has described elsewhere (Macht. Physiol. Zool. **3:** 412, 1930). It was found that cobra neurotoxin, even in doses of 100 mouse units to 200 cc. of water, effected a primary sedation from which the fish gradually recovered and after which they exhibited considerably greater activity. An experiment of 24 hours' duration produced no deleterious effect whatever. Neither was a solution containing 100 mouse units of crude cobra venom in 300 cc. of water toxic for goldfish. Morphine, dilaudid and pantopon, on the contrary, definitely depressed and killed the fish within 12 hours. Morphine was lethal in concentrations of 1:1,500; codeine, in concentrations of 1:2,000; pantopon, in concentration of 1:15,000; dilaudid, in concentrations of 1:5,000; and heroin, in concentrations of 1:12,000. An ichthyometric comparison of cobra venom with two saponins, digitonin, 1:100,000, and saponin, Merck, 1:2,000, revealed that the last two were very toxic for the fish while the venom was not. This observation contradicts the view of those German writers who regard the active principles of cobra venom as chemically related to vegetable saponins. Furthermore, quantitative tests on higher animals which the present writers made of both digitonin and saponin, Merck, for some indication of analgesic and local anesthetic property also yielded negative results.

Pharmacodynamics of para-aminothymol and 2-hydroxybenzylidene-4-aminothymol. DAVID I. MACHT and W. T. SUMMERFORD (by invitation). *Pharmacological Research Laboratory, Hynson, Westcott & Dunning, Inc., Baltimore, Md.* (Read by title.)

These compounds, synthesized by the junior author with a view to determining primarily any latent antipyretic property they might possess, were subjected to a general pharmacological examination. Para-aminothymol hydrochloride is soluble in water but very unstable at room temperature. Intraperitoneal and subcutaneous injections of doses of 10 mgm. in mice had little effect. Doses of 0.5 gram per kilogram of rabbit, administered by stomach tube, were fatal. Smaller doses (0.2 gram) in rabbits caused loss in weight and appetite but did not impair kidney or

liver function. The phytotoxic index of *Lupinus albus* seedlings grown in solutions of 1:10,000 was 80 per cent. Intravenous injections in cats of 20 mgm. per kilo weight depressed the heart and paralyzed the respiration. Even larger doses of the drug produced no antipyresis in either normal rabbits or rabbits with artificial fever.

The compound 2-hydroxybenzylidene-4-aminothymol is insoluble in water and had to be studied either in alcoholic solution or by administration per os. Phytotoxic index of *Lupinus albus* seedlings grown in solutions of 1:20,000 of this drug with one per cent alcohol was 67 per cent. The lethal dose for rabbits administered by stomach tube was 0.25 gram per kilo weight. Small doses, given orally, did not impair kidney or liver function of rabbits. Hydroalcoholic solutions in 5 per cent alcohol in saline, administered intravenously to cats under ether anesthesia, depressed circulation and respiration. No antipyretic effect was observed in either the normal or fevered animals.

The effect of deprivation of substrate on the motility of human spermatozoa. JOHN MACLEOD (introduced by J. C. Hinsey). *Department of Anatomy, Cornell University Medical College, New York City.*[1]

When human spermatozoa are removed from the seminal fluid and suspended in glucose-free Ringer's solution at 38°C., motility falls off progressively until it fails completely at from four to six hours. The complete failure of motility is delayed if the spermatozoa are kept at low oxygen tensions during the period of glucose deprivation. The oxygen tension most conducive to prolonged life of the spermatozoa lies between 2–5 per cent.

After complete failure of motility due to lack of glucose, recovery of motility can be induced by the addition of glucose, fructose, mannose, maltose, or glycogen, but no other substrate yet tried is utilized by the spermatozoa for motility. The recovery of motility under these conditions is dependent upon the oxygen tension to which the cells have been exposed during the period of glucose deprivation. In the range of oxygen tensions between 10 per cent and 95 per cent, the failure of motility is virtually irreversible though isolated cells may show some recovery. Under anaerobic conditions, the recovery is not as complete as that shown when the cells have been exposed to oxygen tensions between 2 per cent and 5 per cent.

Respiratory responses from stimulation of the medulla of the cat. H. W. MAGOUN and LINDSAY E. BEATON (by invitation). *Institute of Neurology, Northwestern University Medical School, Chicago, Ill.*

The respiratory responses elicited by Pitts, Magoun and Ranson (Am. J. Physiol. **126:** 673, 1939) on stimulation of the reticular formation of the medulla of the cat, have been reviewed with reference to the voltage of stimulating current, the distribution of reactive areas, and the effect of lesions at the point of stimulation.

It has been found that by increasing the intensity of stimulation within the range of 1 to 8 volts (8 volts were used in the previous study), the number of reactive points was only slightly increased, while the magnitude

[1] Supported by a grant from the National Committee on Maternal Health.

of responses from points reactive to threshold stimuli became progressively augmented. With 300/sec. condenser discharges, the threshold for inspiratory responses was 1 to 2 volts, that for expiratory reactions 3 to 4 volts. Similar thresholds were found using 60/sec. sine waves.

The presence of a dorsal zone from which expiratory reactions were obtained and a ventral zone from which inspiratory reactions were elicited, within the limits defined by Pitts, Magoun and Ranson, has been repeatedly confirmed.

Variable results were obtained by repeating stimuli in the early minutes after producing lesions around the point of stimulation. These are attributed in part at least, to the balance struck between two opposing factors: a transient depression of excitability of tissue around the lesion, and a transient fall of resistance within the lesion as compared with that of normal brain tissue. In a number of instances a steady state developed within 10 to 15 minutes in which responses were severely impaired or abolished to stimuli below 3 to 5 volts, applied within lesions of less than 1 mm. radius, while less impaired reactions were obtained with greater intensities. Further study of this procedure should be made before using it as a simple test of current spread.

These results do not support the view (J. M. Brookhart, Am. J. Physiol. **129**: 709, 1940) that the respiratory responses in question are dependent upon the activation of large indiscriminately situated regions of the medulla by widely spreading stimuli, but indicate on the contrary, definite areas responsive to locally acting excitation.

The effect of smoking upon respiration. R. J. Main and J. H. Weatherby (by invitation). *Department of Physiology and Pharmacology, Medical College of Virginia, Richmond.*

When studying human respiration and related physiologic functions, it is necessary to know for how long the pharmacologic effects of a cigarette persist, in order to interdict the subjects from smoking for at least that period of time beforehand. In our first attempt to study this effect, we utilized a large rubber bag over chest and abdomen connected to a spirometer, which gives a semi-quantitative value of pulmonary ventilation. Although this method indicated a slight increase immediately following smoking, the values may have been within experimental error.

Consequently alveolar CO_2 determinations were made at frequent intervals before and after smoking, along with blood pressure and pulse rate. An attempt was made to standardize the smoking technic which tended, if anything, to emphasize the effects. The results indicate that there is a distinct depression of alveolar CO_2 tension immediately after smoking a cigarette, but that it returns to approximately normal within ten minutes. Since the effect of a cigarette upon the circulatory system lasts from one-half hour to one hour or more, it seems probable that by the time the blood pressure and pulse return to normal, the effect on respiration has long since disappeared. However, since it does require such a considerable time for the circulation to return to normal, it would appear advisable that smoking be routinely prohibited for two hours previous to beginning a physiologic experiment.

The stimulating effect of smoking upon respiration is probably due directly to a pharmacologic agent in the smoke, such as nicotine, rather than

to any possible secondary effect of the increased blood pressure, since certain vasopressor drugs have no effect upon alveolar CO_2. This agent might stimulate the carotid body, or affect the respiratory center by the production of cerebral ischemia through vasoconstriction.

Electrolyte changes in traumatic shock. J. F. MANERY and D. Y. SOLANDT. *Departments of Biochemistry and Physiology, University of Toronto, Toronto, Canada.*

Thigh muscles of 35 dogs were traumatized by administering many light blows with a rubber mallet. Shock, often accompanied by considerable haemorrhage, usually developed in 3 to 17 hours following this procedure. Ether anaesthesia was used during the traumatization. Blood samples were taken from the external jugular vein in a preanaesthetic period and at intervals until death ensued. Terminal samples were obtained from the carotid artery and the heart.

The changes in the red cell volume, the weight of 1 ml. of plasma, and the water content and chloride concentration of the plasma were small; in some cases there was an indication of dilution. The plasma potassium changes were larger. In the 11 cases when terminal samples were obtained, plasma from the carotid artery showed increases ranging from 18 to 127 per cent; blood from the heart showed higher values. It is a question whether this increase is the cause or the result of the fall in blood pressure. The plasma potassium of samples taken midway between trauma and death were variable, although in general a rise was found. Muscle analyses showed that only those severely injured lost an appreciable amount of potassium. The role of these tissues as a source of the increased plasma potassium will be discussed.

The effect of chemical agents on the "local response" of large single crustacean axons. GEORGE MARMONT (introduced by K. S. Cole). *Wm. G. Kerckhoff Laboratories, California Institute of Technology, Pasadena.*

The local electrical response from isolated, large (80 to 100 μ), single axons, mainly those of the rock lobster, *Panulirus*, was studied. The fibers were held by platinum electrodes immersed in mineral oil, after Hodgkin's method. The preparation was treated with the various chemical agents by running a drop suspended from a pipette along the desired region of the fiber.

It was possible to materially raise the threshold of or abolish the propagated response, and therefore the action potential, while still obtaining a large local response, by treating the axon with one of the following solutions in sea water: KCl, 0.4 per cent; $CaCl_2$, 0.7 per cent; $MgCl_2$, 0.3 per cent; cocaine, 1.0 per cent; and veratrine, one part in one million. The local response from potassium treated fibers was larger than that from untreated ones, for the same stimulus strength. However, the local response disappeared reversibly upon washing the fiber with iso-osmotic KCl. With calcium and magnesium treatment, the subliminal response was smaller than normal. Because the local response was not masked by the action potential, stronger stimulating shocks could be given. A saturated yohimbine solution in sea water also prevented masking of the

local response, since the refractory period of the action potential was increased to more than a second while that of the local response was little affected.

The effect of stimulus strength and the spacial spread of the larger, unmasked local response made possible by treating the axon with potassium, were studied. With adequate stimulus a response of 5 to 10 mv. was obtained several mm. from the cathode. It was found that at any one point along the fiber the latent period of the response tended to remain constant with increasing stimulus. The latent period increased with distance from the cathode but the recovery curves of the local potential at all active points coincided.

The ionic permeability (electrical conductance) of the sensitized nictitating membrane of the cat. ROSE MARRAZZI[1] and AMEDEO S. MARRAZZI (introduced by G. B. Wallace). *Department of Pharmacology, New York University College of Medicine, New York City.*

The nature of the changes responsible for the manifold increase in responsiveness of denervated tissues to agents introduced into the blood stream is unknown. Suggested explanations have fallen into two groups. The first postulates changes only after the exciting agent has reached its site of action; e.g., that once there its greater effectiveness is due to the presence (accumulated during disuse) of an increased quantity of some complimentary substance essential to its action, or that the greater response is due to a deficit of a substance (such as a destructive enzyme) normally limiting the action of exciting agents. The second group attributes the sensitivity to an easier access of the exciting agent into the cell, because of an increased permeability of the cell membrane.

For the cat nictitating membrane the only basis for the theory of increased permeability is the so-called non-specificity of sensitization, since it is observed not only to adrenaline but also to other substances including K^+ and Ca^{++}. The hypothesis then rests on its applicability to all exciting agents including ions. Since the ionic permeability of a tissue may be measured by determining its electrical conductance, or the ease with which a weak, non-stimulating current traverses it, the hypothesis is susceptible to direct test. This was done without necessarily subscribing to the postulate that an agent need traverse the cell membrane to exert its action. The transverse conductance of the isolated muscle of the nictating membrane sensitized by denervation was compared to that of the normal membrane of the other, control eye in a series of cats.

The muscles were removed to a moist chamber immersed in a constant temperature bath and the transverse conductance measured at 1,000 cycles with a Wheatstone bridge utilizing a parallel capacity, and an amplifier in the detector circuit.

The conductance of the sensitized muscles was not consistently higher or lower than that of the controls in the same animals. Electrical measurement does not support the concept that the ionic permeability of the sensitized smooth muscle of the cat's nictitating membrane must be increased. In view of these results impedance measurements have not been extended through a range of frequencies at this time.

[1] Christian A. Herter Fellow.

Modification of the flux equilibrium for bioelectromotive force in the frog's skin. GORDON MARSH and LOREN CARLSON (introduced by J. H. Bodine). *Department of Zoology, State University of Iowa, Iowa City.*

Hydrogen peroxide increases the E.M.F. of frog skin above the values obtained in oxygenated Ringer. The form of the curve of the E.M.F. plotted against peroxide concentration is roughly bell-shaped. This corresponds to the expectation for an oxidation-reduction system at flux equilibrium as the source of the electrical energy as originally formulated by Lund. The effect of peroxide is partially inhibited by cyanide as it should be if the former is operating through an iron catalyst. A measurable antagonism exists between the two in the effect upon the potential; e.g., 3×10^{-4} M cyanide reduced the potential to 41 per cent of the value in oxygenated Ringer's solution, whereas the same concentration in the presence of 1×10^{-4} M H_2O_2 reduced it to 58.5 per cent. No such antagonism could be demonstrated with ethyl urethan, the depression produced by 0.32 M urethan being practically the same in the presence and in the absence of the peroxide. This is to be expected if urethan inhibits a dehydrogenase complex in the skin.

Under the conditions existing in the skin in oxygenated Ringer where the overall velocity of the respiratory process is limited by either the mobilization of oxygen or the activity of some intermediate carrier, the addition of oxidizable substrate in the form of pyruvate (0.001 M) produces no change in the normal potential difference across the skin. The addition of pyruvate in the presence of hydrogen peroxide, (8×10^{-5} M) however, results in an increase in potential (48 per cent) above that which is produced by the peroxide alone (26 per cent). An analogous effect of pyruvate was found at high concentrations of peroxide which normally depress the steady E.M.F. below that found in oxygenated Ringer, pyruvate preventing or reducing the depression. The increase in E.M.F. on increasing the concentration of available oxidizable substrate verifies a specific prediction from the flux equilibrium concept.

Relation of the excitability cycle of the geniculate-striate system to certain problems of monocular and binocular vision. W. H. MARSHALL and S. A. TALBOT. *Laboratory of Physiological Optics, Wilmer Ophthalmological Institute, Johns Hopkins University, Baltimore, Md.* (Read by title.)

The punctate organization of the primary projection system, in which a wide range of reciprocal overlap occurs through an ascending series of synapses, provides a cortical "mosaic" which is finer grained than the retinal mosaic. Physiologically the operation must be dynamic, governed by at least three essential variables: eye movements (physiological nystagmus), modulation of cortical reactions by "spontaneous" activity, and cycles of excitability within the primary projection system.

We have observed the latter factor in the geniculate-striate system in cats and monkeys under chloralose and nembutal anesthesia of such depth that spontaneous activity and random variations in response of the optic nerve to electric stimulus did not embarrass amplitude measurement. The conditioning and test-shocks were applied over repeated cycles under conditions of equilibration. The cycle frequency of $\frac{1}{2}$ to 5 per second somewhat replaces the scanning effect of physiological nystagmus, since components of eye movements range from 1 to 50 per sec.

These data from the cat consistently show a phase of supernormality lasting about 20 msec., followed by a phase of subnormality which peaks at about 50–70 msec. During the supernormal phase spacial recruitment occurs at each synapse, giving favorable conditions for spacial summation by means of cascade amplification through various synaptic levels. The final peaking effect will be greatest where reciprocal overlap is most complete, providing a mechanism for line integration and contour perception which latter in image dimensions approximates $\frac{1}{2}\frac{1}{0}$ cone diameter.

During the supernormal phase spacial summation is at a maximum, providing for line integration; conversely during the subnormal phase spacial recruitment is at a minimum and the channeling of impulses is in a direction to give the retinally limited case for two point discrimination, of approximately one cone diameter.

These factors apply equally well to binocular vision, in which similar interaction cycles are found in the cat and probably are also present in the monkey; though in the latter animal we have not yet separated a supernormal from the subnormal phase.

The relationship between basal metabolism and summated tissue respiration in the dog. A. W. MARTIN and F. A. FUHRMAN (introduced by J. Field, 2d). *Department of Zoology and Physiology and Department of Pharmacology, University of Washington, Seattle.*

It has long been known that in a series of homeotherms the resting metabolism varies inversely with body size. In 1925, Terroine and Roche (Arch. intern. de physiol. **24:** 356, 1925) and Grafe, Reinwein and Singer (Biochem. Ztschr. **165:** 102, 1925) reported that the respiration of homologous tissues in vitro was very nearly constant. According to their work, in animals larger than the mouse, summated tissue respiration should exceed resting metabolism, in the rat by about two-fold, in the dog by five-fold.

In 1939 this hypothesis was tested for the rat by Field, Belding and Martin (J. Cell. and Comp. Physiol. **14:** 143, 1939). It was found that summated tissue respiration was less rather than greater than resting metabolism in this form, and that summated tissue respiration plus a reasponable allowance for minimal functional activity would account for the resting metabolism.

However it was clearly desirable to examine these relations in other forms. This we have now done for the dog. In the average dog in our series (19 animals) summated tissue respiration (extrapolated to the time of death of the animal) will account for 79 per cent of the resting metabolism, while at the time of beginning the measurement of respiration summated tissue respiration amounted to 72 per cent of the resting metabolism. These two values agree rather well with that of Barcroft (Ergebn. d. Physiol. **7:** 699, 1908), which was 82 per cent (organs perfused *in situ*).

When values of resting metabolism of the rat and the dog are compared on a unit weight basis, using the data of Field *et al.* for the former and ours for the latter, the ratio is about 3:1. A similar comparison for excised tissue yields a ratio of about 2.8:1. Thus it appears that in the dog, as in the rat, resting metabolism can be accounted for by summated tissue respiration and minimal functional activity.

The effect of cyanide upon the viability of embryos and adults. ROSEMARY D. C. MARTIN (by invitation) and KENNETH C. FISHER. *Department of Biology, University of Toronto, Toronto, Canada, and the Marine Biological Laboratory, Woods Hole, Mass.*

As indicated by the ability to withstand relatively short immersions in 0.001 M cyanide embryos of squid, salmon, trout and frog become increasingly sensitive to "anaerobiosis" as development proceeds. The over-all change in sensitivity from egg to adult is most marked in the squid and least so in the frog. In all the forms studied, however, the inhibition of respiration by N/1000 NaCN is high and more or less constant throughout development.

In spite of the capacity for anaerobic glycolysis commonly possessed by embryonic tissue, it appears likely that the basis of sensitivity to cyanide (and hence to oxygen lack) is physiological rather than chemical, and closely bound up with the factor of general organization.

In the light of available evidence regarding the ability of lower organisms to withstand anaerobiosis, the present findings may be thought to be indicative of physiological recapitulation.

The distribution of vitamin E in organs and tissues of the rat. KARL E. MASON. *Departments of Anatomy, Vanderbilt University School of Medicine, Nashville, Tenn., and The University of Rochester, School of Medicine and Dentistry, Rochester, N. Y.* (Read by title.)

More than 300 bio-assay tests have been carried out to determine the vitamin E content of various tissues of 1, low E rats, reared on a stock diet providing 3 to 5 times the minimal daily requirement of vitamin E, and of 2, high-E rats, fed a diet containing about 96 per cent fresh raw wheat germ for 2 to 4 months. Tissues were fed in a fresh or frozen state to sterile E-deficient primiparous females during the first 10 days of pregnancy, and the response determined by autopsy or by laparotomy on the 16th day of pregnancy.

The ratio between the number of grams of fresh tissue from low-E and from high-E rats, respectively, required to give a positive response in about 50 per cent of bio-assay tests has been estimated to be approximately as follows: liver 110:8; muscle 65:17; kidney 60:13; body fat 50:17; heart 40:10; lung 35:8; testis > 60:15; epididymis > 40:12; newborn 225:50. These data indicate that, except in the liver, the storage of vitamin E in the tissues increased only 3 to 5 times following wheat germ feeding. Placenta and uterus from high-E rats proved to be as potent as heart and lung, respectively, and mammary gland twice as potent as liver tissue. Less complete data indicate that the spleen, prostate, pancreas and thymus also contain appreciable amounts of the vitamin. As much as 80 grams of blood, and 46 grams of brain and spinal cord, from high-E rats gave no response.

The importance of the pressor receptor nerves in the vasomotor response to gravity.[1] H. S. MAYERSON. *Laboratory of Physiology, School of Medicine, Tulane University of Louisiana, New Orleans.*

Dogs, anesthetized with Na barbital or chloralose, were tilted from the

[1] Aided by a grant from the David Trautman Schwartz Research Fund of Tulane University School of Medicine.

horizontal to the upright, feet down, position. With the change in position there was a sharp drop in intrafemoral and intracarotid blood pressure followed within 10 seconds by a compensatory rise of varying degree. The intrafemoral pressure usually remained from 6 to 20 mm. Hg above the control horizontal level through the F.D. (feet down) period while the intracarotid pressure remained from 10 to 40 mm. Hg below this level. Control observations with the animal in the horizontal position showed no significant blood pressure differences in the two arteries. In the F.D. position the differences were roughly equivalent to the distances of the puncture points from heart level. On return of the animal to the horizontal position there was usually an immediate rise of from 5 to 40 mm. Hg followed within a few seconds by a 5 to 15 mm. Hg dip below and subsequent recovery to the control level.

Denervation of the carotid sinuses or cutting the vagi, thus eliminating the two aortic nerves, uniformly diminished the animal's ability to compensate for gravity. The secondary rise in pressure in the F.D. period as well as the rise after the return to the horizontal position were smaller. In some cases compensation failed during the F.D. position. Elimination of both pressor receptor mechanisms resulted in complete absence of compensation and in many cases in a drop in pressure to shock levels from which recovery was slight. The importance of the pressor receptor nerves in the compensatory response was strikingly illustrated by the changes in intrafemoral pressure. The hydrostatic component present before denervation disappeared and the values in the F.D. position were 30 to 60 per cent lower than the control levels.

The storage of the major liver components. J. J. McBride (by invitation), M. M. Guest (by invitation) and E. L. Scott. *Department of Physiology, College of Physicians and Surgeons, Columbia University, New York City.*

A number of workers have shown that water storage in the liver accompanies glycogen deposition. They have assumed that the weight of the non-glycogen solids remained constant while the liver water and glycogen varied. This assumption is untenable. The data of MacKay and Bergman were recalculated and interpreted to indicate that during glycogenesis induced by feeding pure glucose there is a deposition of as much as 32 per cent of unidentified non-glycogen solids.

In our work the liver weights of male rats of the same age were adjusted to a uniform initial body weight. The content of non-glycogen solids in each experimental group was multiplied by the ratio of non-glycogen solids to water in the control (zero glycogen) animals and the product subtracted from the total water, yielding the water associated with glycogen. "Apparent" ratios of glycogen to water were thus established. It is shown that these "apparent" ratios vary with a change in the non-glycogen solids and that there is a correlation between a low "apparent" ratio and a low percentage of liver water. This correlation can be accounted for in rats fed a stock diet by our finding that glycogenesis is accompanied by an increase of liver fat.

When there is no change in the non-glycogen solids of the liver each gram of glycogen deposited is accompanied by approximately 2.7 grams of water.

Theoretical considerations indicate that this water may be held in hydrate formation.

A comparison of bone ash, modified line test, and radiographic methods for the evaluation of the results of chicken vitamin D assay. EVAN W. MCCHESNEY (by invitation), EDMUND HOMBURGER (by invitation), and DAVID R. CLIMENKO. *Research Laboratories, Winthrop Chemical Company, Inc., Rensselaer, N. Y.*

It is shown for a typical chicken vitamin D assay that the standard bone ash method, the modified line test of McChesney and Homburger, and the radiographic technique of Olsson all lead to substantially the same results for the potency of the preparations tested. The bone ash method has an important advantage over the other two methods in that there is no question of personal judgement involved in the evaluation; however, it is very tedious and time-consuming, and not infrequently the correlation between the bone ash values and the degree of protection from, or cure of, rachitis is unsatisfactory. The radiographic method of evaluation has the advantage of rapidity and the fact that no special preparation of the tissue is required. The modified line test has the dual advantage that it is rapid and that it requires no special equipment. The evaluation is, however, based on less clear-cut criteria than those which have been set up for the standard rat line test.

Locus and nature of crossed inhibition in the spinal monkey. GRAYSON P. MCCOUCH, JOSEPH HUGHES and WINIFRED B. STEWART (by invitation). *Department of Physiology, University of Pennsylvania Medical School, and the Institute of the Pennsylvania Hospital, Philadelphia.*

In spinal cats and dogs crossed inhibition of the ipsilateral flexor reflex is associated with inhibition of the internuncial cord potential of similar degree. In the spinal macaque monkey, on the other hand, a crossed afferent volley confined to alpha, beta, and gamma fibers may markedly inhibit the reflex without modifying the internuncial potential. Stronger crossed stimuli may reduce the internuncial potential to lesser or equal degree than the reduction of the reflex response. With suitable grading of inhibitory and excitatory volleys, the crossed component of the internuncial potential from the excitatory volley proves to be more susceptible to inhibition than the ipsilateral component.

The case in which reflex inhibition occurs without demonstrable change in internuncial potential is of interest in relation to current theories of inhibition. Such theories may be divided into those which refer the reduction in irritability to immediately preceding response of the same units and those which invoke specific inhibitory endings. If absence of demonstrable change in the intermediary cord potential be accepted as evidence that excitatory drive upon the motoneurons is not essentially altered, then it implies that inhibition has occurred in them in the absence of immediately preceding activity.

The mechanism of morphine miosis. F. D. MCCREA, G. S. EADIE and J. E. MORGAN (by invitation). *Department of Physiology and Pharmacology, Duke University School of Medicine, Durham, N. C.*

Morphine inhibits the activity of choline esterase in brain tissue and

blood serum in vitro, Bernheim and Bernheim (1936) and blood serum in vitro, Eadie (1941). This inhibition would result in an increased and prolonged response to stimuli. It thus seemed possible that morphine miosis might be an exaggeration of the normal light reflex due to this mechanism.

Dogs were anesthetized with morphine and ether, the left optic nerve cut behind the eye ball, avoiding, so far as possible, autonomic nerve injury. After recovery the pupillary responses to light of varying intensity were tested before and after doses of morphine 4.5 to 10.0 mgm. per kilogram. Both manual measurements and photo flash photographs were made at 7–10 day intervals for 2–4 months. Standard conditions were maintained in each trial. Unoperated controls were tested in the same way as operated animals.

No miosis was elicitable upon illumination of the operated pupils. At rest the operated pupil was usually slightly larger, (average 1 mm.) than that of the normal eye. After morphine in both series of animals, the pupils, both normal and operated, were moderately constricted in room light and further constricted in light of maximum intensity (standard opthalmoscope) but only occasionally became pin point for more than a few seconds in maximum light.

Miosis resulting from a given intensity of light was greater with the larger doses of morphine, and progressively increased with increased light intensity. The degree of constriction in the operated eye was slightly less (average 0.8 mm.) than that of the normal eye with a given light intensity. Covering the normal eye to exclude room light for periods of 5 minutes both before and after morphine resulted in some mydriasis in the operated eye (average 1.5 mm.). However after morphine the dilation never equaled that of the same non morphinized animal in light of the same intensity.

We conclude that the chief action of morphine is to exaggerate the effect of the normal light reflex, though the evidence does not exclude a very small direct central oculomotor stimulation.

Functional organization and interrelation of cerebral hemispheres in monkey.[1] Warren S. McCulloch and Hugh W. Garol (by invitation). *Laboratory of Neurophysiology, Yale University, School of Medicine, New Haven, Conn.*

By local strychninization of one hemisphere of the monkey (*Macaca mulatta*) and recording electrical activity of both, the map of functional organization has been extended and the directed functional interrelations of the two hemispheres have been explored.

The new map of functional organization shows that the sensory cortex consists of the following physiologically distinct areas: 6, 4s, 4, 1, 2s, and a region behind 2 consisting of 5 and 7. Anterior to these lies 8s and posterior, area 19s, strychninizations whereof produced suppression of electrical activity of the cortex like that obtained from 4s and from the postcentral suppressor strip which is cytoarchitectonically area 2.

An occipital area was found to "fire" into a narrow strip bordering the sulcus interparietalis.

Areas 9 and 18 have significant callosal projections; areas 8s, 4s, 1, 2s,

[1] Aided by a grant from the Fluid Research Funds of the Yale School of Medicine.

and 19s lack them; the remaining areas of the frontal and parietal lobes give rise to them as follows: 1, from 6 to 6, 4 and the postcentral convolution; 2, from 4 to symmetrical foci and only from the junction of A and L4 and from F4; 3, from 7 to symmetrical foci and at least to 5 and 2s; 4, from 5 to symmetrical foci and to 6, 4, 1 and 2s. These confirm Curtis's conclusions except for differences which may be attributable to lack of specificity of direction in the case of electrical stimulation.

Finally, suppression of electrical activity of the cortex from strychninization of 8s, 4s, 2s or 19s is always bilateral.

The effect of diphtheria toxicosis on cardiac reserve. C. H. McDONALD. *Department of Physiology and Pharmacology, University of Arkansas School of Medicine, Little Rock.*

Dramatic failure of the cardiovascular system sometimes terminates a case of diphtheria toxicosis. This failure may occur without warning late in the course of a convalescence which appears to be progressing well. Whether this failure is due to the heart or the peripheral vessels has been argued. Autopsy usually shows surprisingly little pathological change in the heart.

We attempted to estimate the cardiac reserve in the hearts of dogs made acutely ill by the injection of diphtheria toxin. With the coronary and cranial circulation left intact in some cases and only the coronary circulation left in others, we caused the hearts of such dogs to beat against a raised resistance by occluding the thoracic aorta. Comparison of the ability of these hearts to meet such an added load and to recover from repeated additions of such loads with that of the hearts of untreated dogs was made. These hearts appeared able to withstand such strains equally well with untreated hearts. We again encountered numerous cases of complete ventricular arrest reflexly induced in this series of experiments, confirming our observations described in a paper now in press.

Student apparatus for teaching physiology. A. R. McINTYRE, A. L. BENNETT (by invitation) and J. C. BURKE (by invitation). *Department of Physiology and Pharmacology, University of Nebraska College of Medicine, Omaha.* (Demonstration.)

The apparatus presented consists of: 1, a new type long paper kymograph; 2, stimulating box and signal magnets; 3, ether bottle.

All of this equipment was made in the department of Physiology and Pharmacology, University of Nebraska College of Medicine, and we believe has certain advantageous features not ordinarily found in student apparatus.

Plethysmographic determination of capillary blood pressure in man. CHARLES E. McLENNAN[1] (by invitation), MARGARET T. McLENNAN (by invitation) and EUGENE M. LANDIS. *Department of Internal Medicine, University of Virginia, Charlottesville.*

Capillary blood pressure determinations in man have so far been limited to the skin, and usually to the nail fold where the proximity of arterio-

[1] Commonwealth Fund Fellow.

venous anastomoses may alter even direct readings. Because the total volume of the capillary vessels in a segment of tissue is much greater than the volume of the other portions of the vascular tree in the same segment, it seemed possible that the average pressure in the capillary vessels of muscle and skin might be determined plethysmographically by measuring the external pressure required to produce the greatest decrease in the vascular volume of a segment of the forearm.

The pressure plethysmograph already described (Landis and Gibbon, J. Clin. Investigation **12:** 105, 1933) was used to apply pressures of 5 to 90 mm. Hg to the surface of a segment of the forearm. The vascular volume at each pressure was determined by measuring change in volume produced by *a*, sudden compression of the brachial artery, and *b*, by sudden release of the brachial artery after prior compression. Plotting vascular volume against the applied external pressure yielded for each subject a pressure-volume curve, the summit of which was taken to indicate average capillary blood pressure in the tissues of the forearm.

Plethysmographic capillary blood pressures of 20 normal subjects by three somewhat different techniques at 34°C. agreed closely with average values previously obtained by the direct micro-injection method (Landis, Heart **15:** 209, 1930). Capillary blood pressure was elevated by venous congestion, by exercising the muscles of the forearm, or by lowering the forearm below heart level. Elevating the extremity or cooling the forearm to 14°C. reduced capillary blood pressure. Cooling the forearm or venous congestion reduced the changes in vascular volume conspicuously, whereas local heating to 44°C. increased vascular volume somewhat but did not elevate capillary blood pressure in the forearm. Simple reactive hyperemia produced by 2 to 5 minutes' interruption of blood flow increased vascular volume slightly but did not raise capillary blood flow measurably.

Preliminary results of studies on patients with vascular disorders will be described. They indicate that the method may be useful in studying capillary blood pressure in clinical conditions.

A comparison of the basal heat production of identical and fraternal twins.

W. C. McNelly (introduced by F. A. Hitchcock). *Department of Physiology, Miami University, Oxford, O.*

The O_2 consumption of five pairs of identical twins, two pairs of fraternal twins of the same sex, and two pairs of fraternal twins of different sex was measured with the Benedict-Roth apparatus. All determinations were made under basal conditions and both members of a pair of twins were tested on the same morning. The results were calculated in terms of Calories per square meter of body surface per hour. The differences between the members of a pair of identical twins ranged from 0.3 to 0.7 Calorie per square meter per hour; the average difference was 0.46 ± 0.15 Calorie per square meter per hour. Four and five determinations respectively were made on each of the fraternal twins of the same sex. The differences between the members of the pairs varied from 0.8 to 5.5 Calories per square meter per hour; the average difference was 3.75 ± 2.44. Three tests were made on each of the fraternal twins of different sex. The differences between members of the pairs ranged from 6.4 to 8.9 Calories per square meter per hour; the average difference was 7.65 ± 1.25.

A study of the functions of the stomach following pyloric obstruction and gastroenterostomy. Jos. MEDOFF (by invitation), F. NEUWELT, J. PATEDJL (by invitation) and H. NECHELES. *Department of Gastro-Intestinal Research, Michael Reese Hospital, Chicago, Ill.*

The surgical literature offers ample evidence that gastroenterostomy in the human is often followed by gastrojejunal ulcers. If, however, gastroenterostomy is performed on subjects with pyloric stenosis and a low gastric acid secretion, the formation of gastrojejunal ulcer is exceedingly rare. We have attempted to put this experience to an experimental test on dogs. These animals were gastrostomized by stainless metal cannulas, and at a later date pyloric stenosis (30–90 per cent) was produced by cutting away a diamond-shaped segment and closing the opening in a longitudinal direction. Subsequently, a gastroenterostomy was performed in a great number of these animals. Gastric secretion was measured, both basal and following the injection of histamine, and the gastric emptying time was determined fluoroscopically. The emptying time of the stomach was not materially affected by pyloric stenosis or subsequent gastroenterostomy. The secretion of acid was not altered uniformly by the above procedures; showing no change, an elevation, or a decrease in different animals. Gastrojejunal ulcer did not develop in any animal following gastroenterostomy, with or without pyloric stenosis. In a number of dogs a considerable degree of atrophy of the pancreas was found either following pyloric stenosis alone, or gastroenterostomy subsequent to the production of pyloric stenosis. This was not accompanied by diabetes, but the dogs lost weight and could not be maintained in a good nutritional condition.

Gastroenterostomy in the dog with or without pyloric stenosis cannot be relied upon to produce more rapid emptying time or a reduced gastric secretion of acid.

The effect of experimental hydronephrosis on the arterial blood pressure.[1] R. S. MEGIBOW (by invitation) and L. N. KATZ. *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.*

We have shown that following complete or partial occlusion of both ureters, the systemic arterial blood pressure rises to hypertensive levels in the majority of the experimental animals; when occlusion is complete or almost complete hypertension persists until death in uremia, but when occlusion is slight only transient hypertension results.

To investigate the mechanism of such arterial pressure elevation more completely and in an attempt to produce chronic hypertension such as has been reported to occur in some cases of human hydronephrosis, the effects of the following additional procedures have been studied, *a*, unilateral hydronephrosis; *b*, unilateral hydronephrosis and contralateral nephrectomy; *c*, unilateral hydronephrosis and contralateral renal ischemia, and *d*, unilateral hydronephrosis and contralateral renal ischemia followed by removal of the ischemic kidney. As in our previous study, trained dogs were employed, the blood pressure being obtained by the Hamilton manometer. Hydronephrosis was produced either by partial occlusion of the ureter with a Goldblatt clamp or by ureteral-vein anastomosis.

We have been unable to produce chronic hypertension by uncomplicated hydronephrosis. However when unilateral hydronephrosis is combined

[1] Aided by the A. D. Nast Fund for Cardiac Research.

with contralateral renal ischemia, one of three results were obtained, namely, 1, aggravation of pre-existing hypertension; 2, crystallization of a fluctuating into a permanent hypertension, and 3, the appearance of hypertension previously not existent. The changes in blood pressure appear to depend upon two factors consequent to the hydronephrotic process, *a*, a decrease in renal blood flow secondary to the increase in intraureteral pressure and presumably in intratubular pressure, and *b*, a decrease in the amount of normal renal tissue with consequent increase in the ratio of ischemic to normal kidney tissue.

The "leaping" phenomenon: a sign of striatal damage.[1] FRED A. METTLER and CECILIA C. METTLER (by invitation). *Department of Anatomy, University of Georgia School of Medicine.* (Motion picture demonstration.)

The effects following removal of the frontal cortex have been extensively studied. Among these effects the loss of the hopping and placing reactions is significant. If, in addition to the cortex, significant portions of the striatum are removed there is added to the familiar behavior pattern a definite increase in spontaneous activity. This motor release requires a few days to develop but endures for a long period of time. Shortly after operation, cats sustaining striatal injuries also display a distinctive "leaping" phenomenon which may be elicited by holding the animal erect, upon its hind legs, and pushing down gently upon the shoulder girdle whereupon the animal throws the forefeet into extension and springs into the air. This phenomenon is a reliable indication of striatal injury and is part of a general tendency on the part of animals of this type to resist all forms of impressed movements or to fight resistance of any kind. These reactions are not the result of vestibular stimulation and, indeed, reactivity to stimuli of such an origin is reduced though not absent. After striatal injury there is also a relative increase in movements of a slow and automatic type. These are vaguely reminiscent of athetoid movements but are better formed and not so fragmentary.

Influence of blood extracts from normal, goitrous and diabetic persons on the heart rate of the thyroidectomized rat. ARTHUR E. MEYER and EDGAR A. FERGUSON (introduced by Samuel R. M. Reynolds). *Research Laboratory of The Maltine Company and Metabolic Laboratory of Kings County Hospital, Brooklyn, N. Y.*

It has been shown in previous work that fresh thyroid glands, fed to thyroidectomized rats, stimulate the metabolism and heart rate to a variable degree. In feeding quantities equivalent in metabolic effect, the action on the heart was low in normal glands from humans and animals, and in the thyroid from a non-iodized patient with Graves' disease. It was high in glands from iodized toxic goiter patients.

Iodinization of rats and rabbits caused in their thyroids an accumulation of that hypothetic substance responsible for the increase in heart rate in thyroidectomized rats. It was assumed that the iodinization causes a retention in the gland of a substance otherwise released into circulation. In this case the heart stimulating substance should be found in the blood

[1] Financial assistance from the John and Mary R. Markle Foundation is gratefully acknowledged.

at a concentration inverse to its presence in the thyroid. The faculty to stimulate the heart is removed from thyroid material by repeated extraction with alcohol.

Therefore, blood samples from various subjects were dried in warm air and extracted in a Soxhlet apparatus with alcohol. The dried extract was suspended in water and fed to the test rats in three portions for three days. Heart beat and metabolism were determined on the fifth day. From 10–12 cc. of blood were found sufficient for one rat.

No heart stimulation was obtained with the blood of normal persons, of patients with hyperthyroidism but without toxic symptoms, and in Graves' disease after iodine treatment. Positive results were observed in untreated, fully developed Graves' disease, and also in one case with typical toxic symptoms but a metabolic rate of only +6. A single case of Graves' disease remained negative. A case of diabetes, intended as a negative control, turned out positive in various tests but other diabetic blood samples gave negative results. The metabolic rate in all cases was either unchanged or only mildly influenced.

The number of cases studied to the present moment is not large enough to permit a definite conclusion.

A method of irradiating circulating blood in vitro with ultraviolet spectral energy; studies of its physiological effects in vivo application in humans.

G. P. MILEY (introduced by R. Beutner). *Hahnemann Medical College and Hospital of Philadelphia, Philadelphia, Pa.*

A description is given of a Knott irradiation chamber, showing how, because of its construction with a baffle plate, labyrinthine system, enclosed in a metallic, quartz-windowed disk, an optimum turbulency is obtained and the heretofore insurmountable obstacles encountered in irradiating blood with ultraviolet spectral energy, namely, quartz window blood-film formation, and non-penetrability of this energy below a $\frac{1}{2}$ mm. surface layer of blood, have been eliminated in vitro.

The technique of application of this in vitro method to in vivo work is next described, with mention of the source, the wave-length, and the intensity of the ultraviolet spectral energy used, time of exposure to the spectral energy, and amount of blood withdrawn, exposed, and returned to circulation—the amount being calculated according to the formula: $A = KW$, where A is the amount of blood exposed (in cubic centimeters), K is a constant equivalent to 1.5, and W is the body weight in pounds.

A study is presented of the immediate effects, produced in 119 humans by in vivo application of this method, on blood pressure, temperature, pulse and respiratory rates, these individuals initially having blood pressure findings, temperature, pulse and respiratory rates within normal limits. No apparent significant changes have been found.

Studies of the effects of the in vivo application on the hemoglobin content, erythrocyte and leucocyte structure and count of the blood of 215 humans, made 1 to 24 months following initial application of the method show no essential variations, with the exception of an occasional rise in hemoglobin content, erythrocyte and leucocyte count from below normal values to normal limits and an occasional drop in abnormally high white counts to normal values.

Effect of prostigmine and physostigmine on muscular fibrillations. A. T. MILHORAT and T. P. ALMY (by invitation). *Departments of Medicine and Psychiatry, Cornell University Medical College, New York City.*

Prostigmine and physostigmine were administered to patients with chronic progressive poliomyelitis in amounts that decreased the cholinesterase activity of the serum to similar levels in comparable experiments. Significant increase in muscular fibrillations accompanied only slight changes in esterase activity after prostigmine had been given, whereas similar changes in esterase level induced by physostigmine were without effect on the fibrillations. When larger doses of the drugs were used prostigmine had considerable effect on fibrillations, whereas the effect of physostigmine was only moderate. In some patients, prostigmine produced fibrillations in muscles in which no adventitious movements previously had been observed. The muscles of such patients showed increased sensitivity to prostigmine since fibrillations were produced by amounts of drug that had no muscular effects in normal subjects. This phenomenon probably is useful in determining involvement of muscle groups which on neurological examination appear not yet to be affected by the disease. The results suggest that the effect of prostigmine and physostigmine on muscular fibrillations is due only partly to the anti-esterase activity of the drugs and that these drugs have a direct action on skeletal muscle. The findings suggest further that the occurrence of fibrillations in denervated muscle can be explained only in part by the increased sensitivity of the muscle to acetylcholine.

The effects of eserine and acetylcholine on the electrical potential waves of the cerebellar cortex. FREDERICK R. MILLER. *Department of Physiology, University of Western Ontario, London, Canada.*

It was shown by Miller, Stavraky and Woonton (J. Neurophysiol. **3**: 131, 1940) that the application of 1 per cent eserine, followed by 1 per cent acetylcholine (ACh.), to the cerebral cortex evokes large, regular, electrical spike potentials at 8–10/sec. The present report deals with the effects of the same drugs on the small, rapid waves (150–250/sec.) of the cerebellar cortex.

A minute quantity of 1 per cent eserine in saline applied to the anterior lobe of the cerebellum in the decerebrate cat causes an increase in amplitude and frequency of the small, rapid waves; the enlarged waves occur in groups or as scattered spikes; there are also slow potential oscillations. ACh. 1:2000–1:1000 on application increases still further the amplitude and frequency of the fast waves; the augmented waves occur as scattered spikes or in periodic groups; these latter often show a gradual rise and decline as in the groups of waves following faradic stimulation of the cerebellar cortex, as described by Dow (C. R. Soc. Biol., Paris **128**: 538, 1938); the groups are more prolonged and occur at briefer intervals than under eserine alone. ACh. also induces large, slow, random potential oscillations.

The above effects occur immediately and thus depend on local actions of the drugs. There are accompanying changes in tonus, indicating stimulation by the drugs of the cerebellar cortex. The manifestations of eserine and ACh., when in being, are abolished by complete atropinization of the animal; atropinization also prevents the development of the effects of

eserine and ACh. Similar effects are obtained with 1 per cent eserine and 1 per cent ACh. in animals under urethane or chloralose.

The continuous measurement of arterial saturation in man. G. A. Millikan, J. R. Pappenheimer (by invitation), A. J. Rawson (by invitation) and J. E. Hervey (by invitation). *Cornell University Medical College, New York, and the Aero Medical Research Unit, U. S. Army Air Corps, Wright Field, Dayton, O.* (Demonstration.)

A small unit placed over the shell of the ear contains a lamp, two color filters, and two barrier type light-sensitive cells, with which the transmission of either green or red light is measured. The "green" reading depends upon how much total hemoglobin there is between lamp and photocell, and is used as a measure of the degree of vasodilatation or "blood thickness" of the ear. It enables one to choose the correct direct-reading calibration scale for the estimation of arterial oxygen saturation, as measured in the "red" reading. The method has been controlled by van Slyke determinations on eighteen arterial samples, and is found to be accurate to about 5 per cent over the top half of its range and to about 8 per cent over the bottom half.

Vitamin (B fractions) and protein requirements at different environmental temperature levels. C. A. Mills. *University of Cincinnati, Cincinnati, O.*

Using young Wistar rats kept in rooms maintained at 91°F and at 65°F, it has been found that optimal growth response and food consumption require twice as high a dietary thiamine content at 91 than at 65°F. Definite inadequacy at 91°F. prevails up to 0.8 mgm. thiamine per kilo of food, but at 65°F. inadequacy is present only up to 0.4 mgm. per kilo. Pyridoxine (B_6) and pantothenic acid also exhibit higher requirements at 91 than at 65°F. Graphs will be presented to illustrate these findings, and their possible significance in reference to human deficiency diseases and the problems of tropical existence will be discussed.

Repetitive focal discharges and conduction changes related to the induction of ventricular fibrillation.[1] Gordon K. Moe (by invitation) and A. Sidney Harris. *Department of Physiology, Western Reserve University, Medical School, Cleveland, O.*

Further evidence was sought regarding the mechanism by which a brief shock applied during the vulnerable period of the heart cycle induces a *series* of aberrant beats or fibrillation. Simultaneous string galvanometer records of the electrical activity of three different areas on the ventral surface of dog's ventricles were taken by means of contiguous bipolar riding electrodes. Brief rectangular shocks of from 1 to 40 volts were appropriately applied at various moments in the cardiac cycle. Time relations of the resulting aberrant electrical complexes were determined at many points on the cardiac surface by frequent shifting of the riding electrodes.

Measurement of these records shows that of two extrasystoles resulting from a single shock, both originate at the stimulated locus. Isochrons drawn on the heart surface have essentially the same concentric pattern for both the primary and secondary idioventricular beats. Since paired

[1] Aided by a grant from the John and Mary R. Markle Foundation.

extrasystoles result only when the shock is at or near the threshold intensity for fibrillation, and since these paired complexes frequently have the same configuration as the initial complexes in the fibrillation patterns recorded from the same points, it is suggested that fibrillation itself begins with a series of ectopic beats from a single focus.

Analysis of the initial fibrillation complexes before complete dissociation of the several points reveals that such an idioventricular focus usually discharges at progressively decreasing intervals (down to 60–80 msec.), and that the time lag (conduction time) between points in line with, and on the same side of, the stimulating electrodes becomes progressively greater until eventually a point near the focus may undergo its fourth discharge at the same time that a point only 2 cm. distant is activated for the third time. When such a stage is reached, conduction may be completely blocked, resulting in a momentary arrest of the heart, or conduction may be irregularly blocked, leading to fibrillation which begins near the point of stimulation while distant areas of the heart are still being excited rhythmically.

Rôle of the external secretion of the pancreas in the prevention of fatty infiltration of the liver. M. LAURENCE MONTGOMERY (by invitation) and I. L. CHAIKOFF. *Divisions of Surgery and Physiology of the University of California Medical School, San Francisco and Berkeley.* (Demonstration.)

Previous observations have clearly established that raw pancreas contains a factor (or factors), other than insulin, that prevents deposition of fat in the liver. It will be demonstrated by gross specimens, photomicrographs and chemical analyses that the daily ingestion of adequate amounts of pancreatic juice can replace the raw glandular tissue in the prevention of fatty livers in duct-ligated and in completely depancreatized dogs kept alive with insulin. These findings suggest that the maintenance of a normal lipid content in the liver is an essential function of the external secretion of the pancreas.

Fatal blood level of magnesium. R. M. MOORE and W. J. WINGO (by invitation). *Departments of Surgery and Biological Chemistry, University of Texas School of Medicine, Galveston.* (Read by title.)

Although the anesthetic action of magnesium salts (Meltzer and Auer, Am. J. Physiol. **15:** 387, 1906) and their action in lowering the irritability of pain-endings (Moore, Am. J. Physiol. **110:** 191, 1934) have been demonstrated, the concentrations of magnesium ion in the blood necessary for these effects have never been determined. As a preliminary to such a study we have investigated the toxicity of magnesium chloride administered intravenously to cats under ether or nembutal anesthesia. Blood magnesium was determined by a modification of the Hirschfelder and Searles method (J. Biol. Chem. **104:** 635, 1934).

Animals receiving continuous intravenous injection of isotonic magnesium chloride exhibited a moderate respiratory depression which culminated suddenly in fatal respiratory arrest. Caval blood samples taken at death in 13 experiments showed magnesium concentrations from 18.5 to 31.0 mgm. per cent (avg. 24.5). From analysis it appeared that blood level was the lethal factor rather than total dose or injection rate. In 5

additional animals artificial respiration was given while magnesium injection was continued until the heart beat was no longer felt; terminal blood samples showed magnesium values of 47.5 to 72.0 mgm. per cent (avg. 60.3). In another group of 5 animals injection was discontinued prior to respiratory arrest; these animals survived the experiment apparently uninjured although the blood magnesium had been elevated to 20–24 mgm. per cent. It is evident, therefore, that cessation of respiration is the only acute danger from artificial elevation of the blood magnesium.

When a double-isotonic solution combining magnesium and calcium chlorides in isosmotic proportions was injected higher magnesium concentrations were required to kill. In 6 such experiments blood magnesium concentrations at death averaged 65.8 mgm. per cent while blood calcium averaged 36.9 mgm. per cent. In these animals respiration and heart beat appeared to fail simultaneously. Moreover, death was not so sudden as from magnesium alone and cardiac irregularities were noted prior to death.

Central stimulation of respiration during anoxia. CARL A. MOYER[1] and H. K. BEECHER (introduced by R. Gesell). *Surgical Laboratories (Anesthesia), Harvard Medical School, Massachusetts General Hospital, Boston.*

Four dogs lightly anesthetised with evipal after bilateral cervical vagus section and bilateral carotid denervation showed varying degrees of respiratory stimulation when subjected to an oxygen tension of 61 millimeters of mercury.

The stimulation was characterised by a long latent period, a rapid rate and periodicity. During the latent period (3–8 min.) pulmonary ventilation usually decreased slightly and blood pressure fell. When stimulation became well established blood pressure became stabilized or rose slightly and cyanosis decreased.

Readministration of room air was followed by a short period of diminished respiratory exchange which in turn was superseded by a period of hyperventilation differing from the anoxic response in that the amplitude increase was the more prominent component.

A very slight objective increase in the depth of anesthesia abolished the hypoxic hyperpnea but only diminished the post-anoxic hyperventilation. The animals still tolerated the anoxia well.

At a slightly greater depth of anesthesia the usual picture of hypoxia after complete *chemoceptor* and vagal *proprioceptor* denervation was invariably obtained, namely: progressive respiratory and vasomotor depression.

The amount of evipal necessary to annul central low oxygen stimulation is very slight when compared with that necessary to abolish it in the normally innervated animal.

These findings substantiate D'Autrebande's on chronically denervated unanesthetised dogs and fit in well with Gesell's (H)+ ion concept of respiratory control.

The metabolism of the brain in the ketotic state. A. G. MULDER and LATHAN A. CRANDALL, JR. *Department of Physiology, University of Tennessee College of Medicine, Memphis.*

[1] National Research Council Fellow.

The metabolism of the brain in dogs starved and fed on fat alone for from 2 to 6 weeks was investigated by the determination of the arterio-venous differences of ketone bodies, oxygen, carbon dioxide, and glucose. Arterial blood was drawn from the femoral artery while simultaneously venous blood was drawn from the superior sagital sinus. Two of the dogs were not anesthetized during the blood collection; a trephine opening in the skull having been prepared on the day previous to the experiment. The remaining dogs were anesthetized. No differences were observed due to anesthesia.

In eleven of the experiments the total ketone bodies were determined. The level of the ketone bodies in the blood varied from 4.1 to 64 mgm. per 100 cc. (expressed as beta-hydroxybutyric acid). In four of these experiments the arterial blood contained an average of 0.5 mgm. more than that of the venous blood, while in seven experiments the venous blood contained an average of 0.3 mgm. more than did the arterial blood. All of the variations were within the limit of accuracy of the method which is less than 5 per cent. The results indicate no utilization of ketone bodies by the brain.

The respiratory quotient was determined in eight experiments. This ratio varied from 0.95 to 1.07, with one exception; the average R.Q. was 0.98. The average arterio-venous oxygen difference was 8.6 volumes per cent. Glucose was removed by the brain in all of the six experiments in which it was determined, the average utilization being 10.5 mgm. per 100 cc. Determinations of lactic acid showed no appreciable difference between arterial and venous blood in two experiments.

These results demonstrate that the brain of the dog in ketosis uses carbohydrate alone in its metabolism.

Some observations on Brdička's polarographic serum reactions.[1] OTTO H. MÜLLER (introduced by J. C. Hinsey). *Department of Anatomy, Cornell University Medical College, New York City.*

If proteins or their denaturation products are polarographed in a well buffered solution of cobaltous chloride, two well defined waves are observed on the polarogram in addition to the wave due to the reduction of cobaltous ions. This reaction, first observed by Brdička, has been applied by him to the study of cancer and infectious diseases (R. Brdička, Acta Intern. Vereinig. Krebsbekampfung **3:** 13, 1938). It seemed of interest to make similar tests in cases of arthritis and gout. Preliminary experiments have shown that plasma obtained from arthritic patients results in a protein double-wave which is definitely lower than that produced by normal plasma. In the case of gout no significant difference could be established. One difficulty in evaluating these results was the fluctuation in the wave-heights obtained with normal plasma which served as reference. Part of this difficulty could be traced to variations in room temperature. An investigation of the temperature coefficient for this reaction was therefore undertaken over the temperature range of 5–50 degrees Centigrade. The temperature coefficient for the protein double-wave was about 1.7 per cent. This is similar to that of other electro-reductions at the dropping mercury electrode (V. Nejedlý, Collection Czechoslov. Chem. Commun. **1:** 319, 1929). However, unexpected was the observation that the first

[1] Aided by the W. E. Benjamin Fund for the Study of Arthritis.

protein wave had a positive temperature coefficient of about 3 per cent while the second protein wave had a negative temperature coefficient of about 1.3 per cent. This means that with increasing temperature the first protein wave increases at the expense of the second protein wave. Jühling, Tropp and Wöhlisch (Ztschr. Physiol. Chem. **262**: 210, 1939) concluded on the basis of protein denaturations carried out at different temperatures that the first wave is due to a special rearrangement in the protein molecule exposing sulphur groups so that they will react at the electrode. The second wave is considered due to free cystine. Our observation of the temperature coefficients of the two waves seems to throw doubt upon this interpretation.

Studies on the blood of mongrel dogs at high altitude. R. M. MULLIGAN (introduced by R. W. Whitehead). *Department of Pathology, School of Medicine and Hospitals, University of Colorado, Denver.*

Between July 1939 and August 1940 60 normal adult mongrel dogs (39 males and 21 females), ranging in weight between 5 and 19 kilograms, were observed to determine their blood picture. The hemoglobin (Newcomer) values ranged between 11.0 and 18.8 grams with an average of 14.5 ± 1.76 (104.8 per cent Haldane). The red blood corpuscles varied between 5,220,000 and 8,460,000 per cu. mm. with a mean of 6,790,000 ± 700,000. The average mean corpuscular Hb was 21.5 ± 2.45 micromicrograms. The total leucocytes varied between 5,450 and 28,250 with an average of 13,150 ± 5,250. The differential leucocyte count on 200 cells was: Stab neutrophiles—range 0.5–10.0 per cent, average 4.3 ± 2.34; segmented neutrophiles—54.5–91.5 per cent, average 73.9 ± 8.67; lymphocytes—4.5–37.0 per cent, average 17.1 ± 8.25; eosinophiles—0–13 per cent, average—3 ± 2.51; monocytes—0–7 per cent, average 1.7 ± 1.5. Basophiles were never encountered in any of the counts. One normoblast per 200 cells was found in each of three counts.

No relation of hemoglobin, red blood corpuscle count, or mean corpuscular hemoglobin to sex or weight was found.

These results, the first of their kind to be reported from a high altitude (approximately one mile), compare favorably with those done on an almost similar series of mongrel dogs (37 males and 23 females) observed in New Orleans (approximately at sea level) by H. S. Mayerson (Anat. Rec.—47,239, 1930). He found in his series that the hemoglobin averaged 13.5 grams (Newcomer); red blood corpuscle count, 6,160,000 per cu. mm.; total leucocyte count, 11,100 per cu. mm.; neutrophiles, 74.0 per cent; lymphocytes, 20.0 per cent; monocytes, 4.0 per cent, and eosinophiles, 2.0 per cent; basophiles, less than 1 per cent.

Erythrocyte permeability to radioactive potassium. L. J. MULLINS (by invitation), T. R. NOONAN (by invitation), L. F. HAEGE (by invitation) and W. O. FENN. *Department of Physiology, The University of Rochester School of Medicine and Dentistry, Rochester, N. Y.*

The percent penetration of the erythrocytes studied was taken as equal to the potassium radioactivity (KRA, or counts per mol. of K) of the red cells expressed in per cent of the KRA of the plasma, i.e., per cent penetration = $100 \times KRA_{cells}/KRA_{plasma}$. In all cases therefore both cells and plasma had to be analyzed for both total K and for radioactivity.

Thirty-two determinations have been made on human erythrocytes

in vitro. Blood was drawn with various different anticoagulants and incubated with stirring for 10 hours or less at 37°C. after adding 0.1 cc. of isotonic radioactive K to 5 cc. of blood. The samples were equilibrated with various tensions of O_2, CO_2, CO and N_2. Without further experiments it cannot be said that any of these various anticoagulants or gas tensions caused significant differences in the penetration which was about 15% complete in 4 hours and 40% complete in 12 hours.

For experiments *in vivo* the radioactive K*Cl was injected or drunk and blood samples were taken with oxalate at various times thereafter. After centrifuging, cells were either analyzed and counted directly or by the difference between whole blood and plasma values. The approximate times in hours for 30 per cent penetration in a number of different species were as follows: cat, 1.0; rat, 5.0; human, 6.5; rabbit, 7.5; guinea pig, 8.0; and frog, 49.0 (the value for frog cells is extrapolated and it is possible that in these nucleated cells 100 per cent penetration would not be reached). The penetration curves were exponential in nature and it seems likely that the penetration would be complete (100 per cent) if observations could be made at times longer than 24 hours.

Does acetylcholine act specifically as "synaptic transmitter"?[1] D. NACHMANSOHN. *Laboratory of Physiology, Yale University School of Medicine, New Haven, Conn.*

The concentration of cholinesterase is high in strong electric organs (*Torpedo, Gymnotus*) and relatively low in weak electric organs (Ray). If the E.M.F. per centimeter, number of plates per centimeter and concentration of enzyme are compared in the three species, a close parallelism becomes obvious. In recent experiments with Coates and Cox (unpublished) the enzyme concentration was determined in different sections of the electric organ of *Gymnotus* taken successively from the head to the caudal end. The curve obtained was similar to that found by those authors for the E.M.F. per centimeter and number of plates per centimeter. These findings indicate that the discharge is connected with acetylcholine metabolism. This conception is supported by experiments with Feldberg and Fessard where it was demonstrated that nerve stimulation liberates acetylcholine in electric organs and that small amounts of injected acetylcholine produce a discharge, an effect greatly enhanced by eserinization.

Is the correlation between electrical changes and enzyme activity limited to nerve endings? The concentration of cholinesterase rises to particularly high values at synaptic regions but is high everywhere in nerve fibers. Experiments with Couteaux on the superior cervical ganglion after degeneration of preganglionic fibers suggest that the enzyme is concentrated at or near surfaces of nerve fibers, and that the high values at synaptic regions are connected with the increase in surface due to the end arborization. Direct evidence is now offered with Boell for the localization of the enzyme at or near the surface of nerve fibers. The enzyme activity was determined with the Cartesian diver technique separately in sheath and axoplasm of the giant fiber of the squid. Practically all the enzyme of the fiber is located in the sheath, the amount present in the axoplasm is negligible.

These observations suggest—in modification of the original theories of

[1] Aided by a grant from the Dazian Foundation.

Loewi and Dale—that acetylcholine does not act specifically as "synaptic transmitter" of nerve impulses to effector organs, but is intrinsically connected with the electrical changes occurring everywhere at the surface of neurons during activity.

The formation of the R wave of the electrocardiogram.[1] L. H. NAHUM, H. E. HOFF and W. KAUFMANN (by invitation). *Laboratory of Physiology, Yale University School of Medicine, New Haven, Conn.*

Previous studies (Am. J. Physiol. **131:** 687, 1941) indicate that the normal electrocardiogram is formed by the algebraic summation of the action potentials from the right and left ventricles (dextro- and levocardiogram). Summation of the initial portions of these two components yields the QRS complex. Its ascending limb arises from the upstroke of the dextrocardiogram, and its descending limb is formed by the equalization of potential consequent to the onset of the levocardiogram, of which the initial component is directed downward. Two further series of experimenta are here reported in confirmation of this hypothesis. (i) If the interval between activation of the right and left ventricles is lengthened the R wave should be increased in amplitude, because more of the dextrocardiogram would have developed before its neutralization by the levocardiogram. Furthermore, shortening this interval should diminish the amplitude of R. These effects were produced experimentally by warming or cooling one or the other ventricle for several minutes, sufficient to affect conduction to the surface of each ventricle. Warming the left ventricle diminished the height of R, while cooling it increased the amplitude. Warming the right ventricle increased the amplitude of R and cooling decreased it. (ii) If initial deflection of the right ventricular action potential is upward, and that of the left ventricle downward, extrasystoles elicited from the right ventricle should have an upright initial deflection, and those from the left ventricle a downward initial deflection. This is confirmed experimentally when care is taken that the point of stimulation is equidistant from the septal boundary, so that the greater part of the ventricle is activated before there is spread to the other. With these precautions, right ventricular extrasystoles show an upright initial deflection in all three conventional leads, and left ventricular extrasystoles show a downward initial deflection in these leads.

Gastrointestinal secretions during shock. H. NECHELES and WM. H. OLSON (by invitation). *Department of Gastro-Intestinal Research, Michael Reese Hospital, Chicago, Ill.*

Gastro-intestinal secretions may play an important role during the condition of shock, in view of the deranged distribution of fluids within and outside of the circulatory system, and in view of the occurrence of gastro-intestinal ulcers following shock. Therefore, the secretion of saliva, gastric and pancreatic juice, bile and urine, and gastric motility was studied in anesthetized animals before and following shock from trauma or burns. Following both forms of shock, salivary, pancreatic, biliary, and urinary secretions were found to be considerably diminished. Gastric secretion was not affected by traumatic shock, but following burns a rapid and sustained increase of gastric secretion, amounting to several hundred per

[1] Aided by a grant from the Fluid Research Fund, Yale University School of Medicine.

cent, and greatly increased gastric motility, was observed. Salivary, biliary, and pancreatic secretions stimulated by pilocarpine, decholin or secretin, were not effected by either type of shock. The great increase in gastric secretion and motility following burns is the first experimental evidence produced of a disturbance in the mechanism of the stomach following burns. This fact, together with the diminished secretion of bile and pancreatic juice brings forth new light on the problem of Curling's ulcer.

The influence of fasting and of insulin on the concentration of acid soluble phosphorus in the liver of rats. NORTON NELSON (by invitation), S. RAPOPORT (by invitation), GEORGE M. GUEST (by invitation) and I. ARTHUR MIRSKY. *The May Institute for Medical Research, The Jewish Hospital, and The Children's Hospital Research Foundation, Cincinnati, O.*

The concentrations of several fractions of acid-soluble P in the livers of rats were studied following feeding and fasting and after the administration of insulin. The rats had been fed a standard laboratory ration and ranged in weight from 175 to 275 grams. Analyses included the inorganic P, the organic acid-soluble P hydrolyzable by N acid in 8 minutes, the total acid-soluble P, and glycogen in the liver and the blood sugar.

After fasting 15 to 20 hours the concentration of liver glycogen decreased to less than 0.3 gram per 100 grams and the blood sugar fell from 116 (average in fed animals) to 84 mgm. per 100 cc. The inorganic P in the liver increased from 18 (average in fed animals) to 22 mgm. per 100 grams, and the organic acid-soluble P decreased, while the total acid-soluble P did not differ significantly from the average of 100 mgm. per 100 grams found in the fed animals.

One hour after the subcutaneous injection of 2 to 5 units of insulin per animal the concentration of total acid-soluble P in the liver was found to be higher, 112 and 117 mgm. per 100 grams in the fasted and fed rats respectively. The increase was mainly accounted for in the organic fractions although the concentration of inorganic P also was somewhat increased.

Growth of the mammary gland following local application of estrogenic hormone. WARREN O. NELSON. *Department of Anatomy, Wayne University, College of Medicine, Detroit, Mich.* (Read by title.)

The theory that the ovarian hormones stimulate growth of the mammary glands only through their action on the anterior hypophysis and the production therein of one or more mammogenic hormones is controverted by experiments in which application of estrin to the area of one gland has induced growth of that gland only (women, monkeys, rabbits). The present work deals with similar studies in the guinea pig.

Seven gonadectomized male and female guinea pigs received applications of estrin to one mammary gland area. In each animal the estrone dissolved in seasame oil was applied to one nipple and areola. Very small quantities were applied and massaged gently into the skin. Each animal received approximately 3 i.u. daily for 15, 25 and 35 days. In three animals the second nipple was massaged daily with seasame oil alone.

Growth of the nipple on the side receiving applications of estrin was evident by the 8th day and continued throughout the period of treatment

in each animal. In only one instance was any growth observed in the nipple on the side receiving seasame oil only. The mammary glands recovered from the side treated with estrone showed development of ducts and end buds at 15 days, and progressive growth of both ducts and alveoli at 25 and 35 days. A slight growth of ducts was produced in one gland which received applications of oil only.

These results present further evidence that local application of small amounts of estrin to the area of a mammary gland will stimulate growth of that gland only.

The occurrence of hypophyseal tumors in rats under treatment with diethyl-stilbestrol. WARREN O. NELSON. *Department of Anatomy, Wayne University, College of Medicine, Detroit, Mich.*

A study has been made of 93 rats treated daily with varying amounts of diethyl-stilbestrol for periods up to 14 months. Castrated and normal animals of both sexes have been studied and compared with untreated controls. The effects of the synthetic estrogen have been in general comparable to those obtained with similar amounts of naturally occurring estrogens injected over similar periods of time, viz., enlargement of the pituitary and correction of castration changes, stimulation of mammary glands, primary stimulation of corpora lutea with ultimate inhibition of ovaries, enlargement of adrenals, testicular damage, and loss of body weight.

Interest at this time centers chiefly upon some of the effects seen in a series of 28 normal males and females treated with 50 micrograms daily for 8 months or longer. Animals surviving for this period were stunted, showed loss of body hair, and exhibited progressive debility. The enlargement of the hypophysis, which was shown by all animals treated for shorter periods, continued and was markedly accelerated after the 8th month of treatment. At 8 months the average weight of the hypophysis was 75 mgm., at 10 and 11 months, 150 mgm., at 12 months, 210 mgm., and at 13 and 14 months, 325 mgm. No animals have been maintained for longer periods since in each instance a peculiar syndrome developed and lead to a rapid decline. Voluntary ataxy, choreoid movements, spastic weakness, nystagmus, exophthalmos, visual disturbances and defective postural fixation were seen in nine animals during the 13th and 14th months of treatment. In each of these instances an enormous tumor filled the hypophyseal region and encroached upon the surrounding regions of the brain. The tumors were reddish in color, usually bilobate, and were of a semigelatinous consistency. A firm capsule was always present. Microscopically, the tumors could be identified as chromophobic adenomata. Areas of marked vascular dilatation were wide-spread and definitive chromophilic cells were much reduced in both number and size. Many of the chromophobes were very large and the entire architectural pattern of the anterior lobe showed marked aberrations. Posterior and intermediate lobes were small and in some instances ill-defined.

The tubular reabsorption of urea, thiourea and derivatives of thiourea in the dog kidney. H. J. NICHOLES (by invitation) and R. C. HERRIN. *University of Wisconsin, Madison.*

The renal plasma clearance of each of the following compounds, thio-

urea, methylthiourea, phenylthiourea or s-diethylthiourea, was determined with the simultaneous urea and creatinine clearances. 82 to 98.8 per cent of the s-diethylthiourea, 55 to 95 per cent of the phenylthiourea, 50 to 70 per cent of the methylthiourea, 23 to 59 per cent of the thiourea, and 25 to 60 per cent of the urea filtered at the glomeruli were reabsorbed in the uriniferous tubule. At any particular per cent reabsorption of water the per cent reabsorption of s-diethylthiourea was greatest, that of phenylthiourea second, that of methylthiourea third, and those of thiourea and urea the least. The mean ratio of the per cent reabsorption of thiourea to that of urea was 0.987 for 33 periods. The order of tubular reabsorption of these compounds corresponds to their order of relative lipoid solubility. S-diethylthiourea and phenylthiourea are, respectively, 2.6 and 1.4 times more soluble in ether than in water, whereas, methylthiourea, thiourea and urea are, respectively, 49, 116 and 8000 times more soluble in water than in ether.

There was a pronounced linear increase in the percent reabsorption of s-diethylthiourea and phenylthiourea as the per cent reabsorption of water increased. This definite linear increase suggests that these two compounds may be passively reabsorbed in the same region of the tubule as that where most of the water reabsorption occurs. The per cent reabsorption of urea, thiourea and methylthiourea often increased moderately as the per cent reabsorption of water increased, although frequent, irregular changes in the reabsorption of urea and thiourea, or of urea and methylthiourea, suggest that other factors than the per cent reabsorption of water may regulate the reabsorption of these three very water-soluble compounds.

These observations suggest that the passive return to the circulation of certain lipoid-insoluble waste products, such as urea, is resisted because of the lipoid nature of a boundary in each uriniferous tubule cell.

The influence of adrenal cortical deficiency upon the fifth stage of neuromuscular transmission. HAYDEN C. NICHOLSON and W. Y. TAKAHASHI (by invitation). *Departments of Physiology, Harvard Medical School, Boston, Mass., and the University of Michigan, Ann Arbor.*

These experiments were performed upon cats anaesthetized with dial (Ciba), urethane, or pentobarbital. The muscles studied were those attaching to the tendon of Achilles. The leg was fixed by drills inserted into the tibia. The contractions of the muscles were recorded on a kymograph by connecting the tendon to a myographic lever so arranged that the muscles pulled against one or more heavy rubber bands. After central section of the sciatic, the popliteal nerve was stimulated by slightly supermaximal shocks at a frequency of 60 per second.

In a series of seventeen control experiments upon normal cats the neuromuscular response was exactly similar to that described by Rosenblueth and Luco, the late rise of tension characteristic of the fifth stage of neuromuscular transmission invariably appearing. A second series of experiments was performed upon cats which had been bilaterally adrenalectomized from 24 to 48 hours previously. In one cat adrenalectomized 24 hours before the beginning of the experiment there developed a fifth stage of normal height but not as well maintained as normally. In another cat adrenalectomized 24 hours before the beginning of the experiment no fifth stage appeared. In one experiment performed 48 hours after adrenal-

ectomy a fifth stage appeared but the tension developed amounted only to about 6 per cent of the normal. There have been three experiments performed 48 hours after adrenalectomy in which the animals remained in relatively good condition well beyond the maximum time of appearance of the fifth stage in the control series. In none of these did a fifth stage develop, although the muscles still responded to direct stimulation. In all the experiments upon adrenalectomized animals the height of the initial contraction and the rapidity of the decline to the fatigue level were within normal limits. A cat which forty-eight hours previously had been subjected to a sham operation developed a fifth stage normal in every respect.

The distribution of injected radioactive potassium in rabbits and other animals. T. R. NOONAN (by invitation), L. J. MULLINS (by invitation), L. F. HAEGE (by invitation) and W. O. FENN. *Department of Physiology, School of Medicine and Dentistry, The University of Rochester, Rochester, N. Y.*

Using the artificially radioactive isotope, K^{42}, as a tracer element, studies on potassium distribution have been made in rabbits, frogs, humans and cats. In general, the results confirm those found and reported in rats.

In rabbits, killed at varying times after subcutaneous injection of radioactive K, it was found that the "potassium radioactivity" (KRA) which is defined as:

$$\left(\frac{\text{Counts per kgm. of tissue} \times 100}{\text{Counts injected per kgm. body weight}} \div \text{mM of K per kgm. of tissue}\right)$$

varied in different tissues as follows:

In plasma—rapid rise to maximum almost immediately after injection follows by slow exponential type of fall;

In brain, sciatic nerve, and erythrocytes—very slow exponential rise, not reaching the plasma value even after 24 hours;

In muscle (leg) and skin—slow exponential rise reaching value equal to plasma in 24 hours;

In liver, heart, kidney, diaphragm, and lung—rapid rise, to equal or exceed plasma in 1 hour, slower falling off to reach values essentially equal to plasma.

In anesthetized rabbits, successive samples of hepatic tissue with simultaneously collected plasma were taken at intervals during and after intravenous injection of radioactive KCl solutions. From the values of KRA of these tissues, it could be shown that radioactive potassium enters the liver cells by some process other than cation exchange, probably by absorption as a salt in approximately isotonic solution.

Data from similar experiments with frogs also showed that most cells are freely permeable to potassium, the exceptions, erythrocytes and ovary, showing slower penetration.

The total exchanged potassium of the body at any time can be computed from the KRA of the plasma, or presumably from the KRA of urine, using the relationship.

$$\text{Total exchanged potassium} \quad \frac{100}{\text{KRA}_{\text{plasma}}}.$$

The average values for the amount of exchanged potassium in mM per kgm. body weight 12 hours after injection in various animals were approximately: rats, 70; cats, 70; rabbits, 50; man, 40. The total potassium found by analysis of the entire animals is as follows: rat, 66; cat, 78; rabbit, 83.

The effect of anoxia on the absorption of glucose and glycine from the small intestine. DAVID W. NORTHUP and EDWARD J. VAN LIERE. *Department of Physiology, West Virginia University Medical School, Morgantown.*

The absorption of glucose and glycine from isotonic solutions in the small intestine of dogs at various reduced oxygen tensions was studied, using Moreau loops. A low pressure chamber was employed to obtain the reduced pressures; oxygen tensions (besides the controls at atmospheric pressure) used were 117, 94, 80, 63 and 53 mm. Hg, the last corresponding to an altitude of 28,000 ft. The dogs were under barbital anesthesia. At least eight animals were exposed to each O_2 tension with each substance; up to 25 in cases where the significance of the result was in doubt. Over 50 control experiments were performed for each substance.

It was found that anoxia up to and including 53 mm. Hg O_2 tension does not significantly alter the absorption of glucose from the small intestine, but that the absorption of glycine was significantly depressed at 53 mm. Hg. It was not, however, changed at any greater O_2 tension. The results indicate that oxidative processes are of more importance in the absorption of glycine than is the case with glucose.

Hypophysectomy in hypertensive rats. ERIC OGDEN (by invitation), ERNEST W. PAGE (by invitation) and EVELYN ANDERSON. *Division of Physiology and the Institute of Experimental Biology, University of California, Berkeley.*

The relationship of the pituitary body to renal hypertension is not quite clear even though Page and Sweet (1937) showed a fall in blood pressure in Goldblatt dogs after hypophysectomy and Griffith and Ingle (1940) showed that hypertension sometimes occurred in rats after removal of the posterior lobe of the pituitary and subtotal nephrectomy.

Rats were made hypertensive by partial ligation of one renal artery and when the blood pressure, measured by the tail plethysmograph, had stabilised, the posterior lobes of their hypophyses were ablated. In those animals which showed *diabetes insipidus*, the blood pressure declined slowly or not at all.

In some animals extensive damage to the anterior lobe was presumed to have occurred on the evidence of no *diabetes insipidus*, loss of weight and testicular atrophy. In these animals there was a complete and immediate decline of blood pressure to the levels normal for non-hypertensive animals.

A number of post-hypophysectomised rats showing well marked *diabetes insipidus* were subjected to partial ligation of the renal artery. The proportion of these which developed hypertension was the same as that expected from normal rats.

In spite of the finding of Ogden, Brown and Page (1940) and others that renal hypertensive animals are hypersensitive to the pressor activity of

posterior lobe extract, this lobe does not appear to be necessary for the development or maintenance of renal hypertension. The anterior lobe, on the other hand, appears to be essential for this phenomenon and a further study of its role which may help to elucidate the mechanism of experimental renal hypertension is in progress.

Thanks are due to the John and Mary Markle Foundation for financial assistance in this work.

Changes in lens shape during stimulation of the cervical sympathetic nerve. J. M. D. Olmsted and Meredith W. Morgan, Jr. (by invitation). *Division of Physiology, Medical School, University of California, Berkeley.*

Photographs of the Purkinje-Sanson images reflected from the eye of an iridectomized cat taken before and during stimulation of the cervical sympathetic nerve show that during such stimulation the image from the anterior surface of the lens is shifted by flattening of this lens surface. Photographs of the profile of the lens under these circumstances afford direct proof that the anterior surface of the lens is flattened during cervical sympathetic stimulation.

Simple modification of the Hanike-Gibbs drop recorder. Wm. H. Olson (by invitation) and H. Necheles. *Department of Gastro-Intestinal Research, Michael Reese Hospital, Chicago, Ill.* (Demonstration.)

The Hanike-Gibbs instrument for recording drops of secretion applies the principle of displacement of an electrolyte solution with constant viscosity and conductivity. The instrument reported by Gibbs in 1927 consists of 5 different parts with a number of connections and does not lend itself easily to experiments in which the rate of a number of different secretions must be measured at the same time. We have therefore modified the instrument, using the same principle as Hanike and Gibbs did. The upper part of two cylinders is solidly connected by a glass tube. The first cylinder is empty and connected to the cannulated duct of an organ, the secretion of which is to be measured. The second cylinder is filled with a 10 per cent solution of sodium citrate. The lower pole of this cylinder has a fine opening through which a drop of the citrate solution is forced when a drop of secretion displaces air in the first cylinder. The falling citrate drop passes between a platinum wire and a brass ring fixed in a glass tube, to which slight constant water suction is applied so that the drop of citrate passes rapidly between the contacts. The contacts are connected in series with 115 volts A.C. or D.C. and a small recording signal magnet. A fuse is introduced into the circuit. Due to the constant suction the amount of current transmitted is small enough to permit the use of an ordinary six volt laboratory signal magnet. The instrument can be made in the laboratory at low cost. We have used it for one year and have found it so reliable, easy to use and maintain that we feel it would be useful to many other laboratories. The instrument must be protected from radiating heat which will expand the air and citrate solution in the two chambers. The instrument can be emptied and refilled respectively within a short time and will work for many hours unsupervised. We have used a set of 4 instruments to record simultaneously salivary, pancreatic, biliary and urinary secretions.

Circulation time in shock. WM. H. OLSON (by invitation), F. NEUWELT, H. GUTMANN (by invitation), H. NECHELES and S. O. LEVINSON (by invitation). *S. Deutsch Serum Center and the Department of Gastro-Intestinal Research, Michael Reese Hospital, Chicago, Ill.*

During shock circulating time is increased, due to progressive failure of circulation, diminution of circulating volume, and increased viscosity of the blood. In order to obtain more information about the correlation between circulating time and other important changes, experiments were performed on anesthetized dogs following spontaneous, traumatic and morphine shock. Circulating time was determined by the cyanide method, and recorded by kymograph, which is necessary for accurate results. The disadvantages of the method will be discussed.

Results. During shock, circulating time usually is increased from 100 to 300 per cent. As a rule, a close correlation between blood pressure and circulating time was found to exist. That is, a drop of blood pressure of more than 20 mm. mercury was in most cases reflected by a corresponding slowing of circulation time and vice versa. Another close correlation was found between circulating time and hemoglobin values. In most experiments, increased hemoglobin concentration as expressed by Sahli or hematocrit values was paralleled by a prolongation of circulating time. At times, slight changes in blood pressure were accompanied by marked changes in hemoglobin concentration and prolonged circulating time. At times, severe changes of blood pressure occurred with little change in hemoglobin concentration, but there was a great prolongation of circulating time. It may be stated, that there is an approximate correlation between the product of the changes in hemoglobin concentration and blood pressure, and circulating time. As explained above, a great change in either one of the two factors, that is blood pressure or hemoconcentration and a small change in the other one and vice versa, would affect circulating time.

Circulating time was affected regularly by the infusion of saline, serum or plasma. The change reflected the efficacy of the fluid used, that is following saline infusion, shortening of circulating time was of short duration only, and following infusion with serum or plasma a prolonged improvement of circulation was observed. These effects were also reflected by the changes in hemoconcentration following the use of the three fluids.

Effect of cortical lesions upon discrimination of direction. M. J. OPPENHEIMER (by invitation) and E. A. SPIEGEL.[1] *Department of Physiology and Department of Experimental Neurology, D. J. McCarthy Foundation, Temple University, School of Medicine, Philadelphia, Pa.* (Read by title.)

Using a previously described method (Spiegel and Oppenheimer, Am. J. Physiol. **125:** 265, 1939) of developing conditioned reactions to angular acceleration and discrimination of direction of accelerated rotation in dogs, the effect of cortical lesions upon the retention of these reactions was studied. A comparison of these experiments with a former series on acquisition of these conditioned reactions on dogs with cortical lobectomies (Oppenheimer and Spiegel, Proc. Soc. Exper. Biol. **45:** 418, 1940) gives the following results.

Ablation of the frontal, parietal or temporal lobes does not definitely

[1] Aided by a grant of the National Research Council to E. S.

impair the acquisition of these reactions nor their retention for three weeks after the cortical operation. Cortical ablations may, however, impair the acquisition as well as the retention of discrimination of direction of rotation at low acceleration. This discrimination was recognized in experiments on labyrinthectomized dogs as an important criterion of the analyzing function of the labyrinth. While the acquisition of discrimination of direction is delayed particularly after bilateral temporal lobectomies, the retention of this discrimination is impaired by ablation of the frontal as well as of the temporal lobes.

Protection from cyclopropane-adrenalin irregularities by various drugs. O. S. Orth and C. R. Allen (introduced by W. J. Meek). *Department of Physiology, University of Wisconsin Medical School, Madison.*

In the dog during cyclopropane anesthesia the heart is so sensitized that its usual response to a standard dose of adrenalin (0.01 mgm./kgm.) is multifocal ventricular tachycardia; whereas, the same dosage administered to the unanesthetized, unrestrained animal as a control test never causes such tachycardia. It is believed that the cardiac sensitization is due to impulses over the thoraco-lumbar sympathetics from a sensitized (or anesthetically unsuppressed) hypothalamic center. Studies have been made to determine procedures which might be used to prevent such stimulation.

Procaine hydrochloride 16 mgm./kgm. administered with the standard adrenalin dosage prevented ventricular tachycardia. As measured by failure to obtain ventricular tachycardia upon succeeding injections of adrenalin, protection was present for not more than twenty minutes following the procaine injection. Smaller procaine doses (8 and 12 mgm./kgm.) failed to protect. Quinidine sulfate 15 mgm./kgm. administered intravenously 10–15 minutes before the standard adrenalin test gave complete protection and this persisted for at least two hours. Other methods of quinidine administration, even to higher total doses, were ineffective. Carbon dioxide in excess of 20 per cent in the anesthetic mixture gave complete protection from ventricular tachycardia but lower percentages (5, 10 and 15 per cent) while no worse than the results when there was no CO_2 in the cyclopropane, gave no evidence of protection. All of the above compounds probably acted by direct myocardial depression.

Ergotamine tartrate $\frac{1}{6}$ to $\frac{1}{8}$ mgm./kgm.; yohimbine hydrochloride $\frac{1}{5}$ mgm./kgm. and F883 (diethyl-aminomethyl-benzodioxane) 2.0 mgm./kgm. also gave complete protection. It is believed that these three drugs acted by blocking the sympathetic impulses from the active hypothalamic center to the heart.

Since smaller doses of quinidine and procaine protect against chloroform-adrenalin irregularities it is believed that the present tests offer further proof of greater sensitization of the heart by cyclopropane than by chloroform.

Effect of body temperature on pancreatic secretion. S. L. Osborne (by invitation) and Harry Greengard. *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

The established therapeutic use of artificial fever and the more recent

trials of artificially induced subnormal temperatures have made pertinent an investigation of the effect of these procedures on the secretions of the body fluids. Therefore the alterations in pancreatic secretion produced by these means have been studied. Anesthetized dogs were given secretin at a constant rate in order to elicit a continuous and regular secretion. In one series of animals the body temperature was raised by means of short-wave diathermy, and in a second series it was lowered, by immersion of the animals in a bath through which cold water was circulated. Raising the body temperature resulted in an increased flow of pancreatic juice up to seven times the original rate, and lowering of the temperature operated to cause complete cessation of secretion. These findings are probably accounted for on the basis of alterations in circulation through the pancreas.

Ovarian irradiation and mammary cancer.[1] SEWARD E. OWEN. *The Cancer Research Unit, Veterans' Administration, Hines, Ill.*

Irradiation to the ovarian area of animals, in amounts sufficient to suppress fecundation has been termed the "castration dose". In mice this dose is about one hundred and fifty roentgens. Early ovariectomy of breast cancer strain mice results in a greatly decreased subsequent incidence of mammary tumors. We find administration of the fecundation suppression dose of X-ray does not decrease subsequent development of mammary

Incidence of breast tumors in irradiated mice (strain C_3H, females)

GROUP	NUMBER OF MICE	NUMBER OF TUMORS	AVERAGE AGE	PERCENT TUMORS	r DOSE	DOSES	TOTAL r
			mo.				
I	9	3	9.3	33	100-D′	1	100
	4	2	9.4	50	51-S′	2	102
	10	4	9.6	40	175-D′	1	175
Total.....	23	9		39			
II	6	1	9.6	16	100.5-S′	4	402
	6	1	9.4	16	500-D′	1	500
	9	1	9.7	11	201-S′	4	804
Total.....	21	3		14			
III	26	17	9.7	65	None		
IV	25	1	9.6	4	None		
V	10	0	4.0	0	None		

D′, indicates deep x-ray; S′, superficial x-ray. I, low dosage group; II, high dosage group; III, non-irradiated controls; IV, non-bred controls, no x-ray; V, histological controls.

cancer as well as ovariectomy. Ovarian irradiation below the fecundation suppression amounts decrease subsequent incidence of breast tumors even though litters are raised following treatment, thus partial ovarian suppression is lasting. Breast tumor incidence was approximately inversely proportional to the x-ray dosages given. Irradiation was given at age four months (plus or minus two weeks) after the mice had raised one litter. Histological controls (V) showed the mice to be non-cancerous at time of

[1] Published under provisions of R. & P. 6727, Veterans Administration.

treatment. Standards for the deep x-ray were 140 KV., 20 ma., 1 mm. Al and 0.25 mm. Cu. plus 3 mm. basswood, T.S.D. 50 cm., rate 17 r./min For superficial x-ray (S′) 100 KV., 5 ma., 3 mm. basswood, T.S.D. 20 cm., rate 201 r./min. Lapse of one day between repeated doses. Area irradiated was the ventral surfaces of the animals. Numbers of animals in the final groups in each series are perhaps too small for exact quantitative analysis but the trend is clear.

Pharmacologic effects of a commercial meat extract on the isolated frog heart. NELLO PACE (introduced by R. J. Main). *Department of Physiology and Pharmacology, Medical College of Virginia, Richmond.*

Frog hearts, cannulated through the sinus venosus, were perfused with cold-blooded Ringer's solution for at least 30 minutes at room temperature, or until the preparation had stabilized. The heart rate and amplitude of contraction were measured at frequent intervals. When the heart was stabilized, perfusion fluid containing the meat extract was substituted for Ringer's solution. Observations were made for one hour or more and finally Ringer's solution was perfused once again for at least another 30 minutes in order to determine again the normal rate and amplitude.

The experimental solutions were made up so that, as nearly as possible, only one factor was varied at a time. The pH and ion concentrations were adjusted to the physiologic range in all cases. Owing to the preponderance of potassium ion in meat extract, it was not necessary to add potassium to most dilutions of meat extract used. The upper dilution limit of meat extract is determined by the potassium present and was found to be a 0.50 per cent solution of meat extract. This corresponds to 4×10^{-3} M potassium ion.

Greater dilutions (to 0.05 per cent) of meat extract show a marked stimulation of amplitude of contraction (10 to 55 per cent increase in 26 hearts) which persists for at least two hours. The rate is apparently little affected. The active substance was found to be dialyzable, heat-stable, soluble in 70 per cent alcohol, and to possess a negative charge at pH 5.6 but not at pH 2.0. Meat extract autoclaved at pH 2.0 no longer shows this effect.

Frog hearts were perfused with substances known to be present in meat extracts in an attempt to duplicate the increase in amplitude. Certain members of the guanidine group gave results most comparable to that of meat extract.

Manifestations of oxygen poisoning in dogs confined in atmospheres of 80 to 100 per cent oxygen. JOHN RANDOLPH PAINE (by invitation), ANCEL KEYS and DAVID LYNN (by invitation). *Department of Surgery and the Laboratory of Physiological Hygiene, University of Minnesota Medical School, Minneapolis.*

Dogs maintained in 99 to 100 per cent oxygen developed respiratory distress within 48 hours and died in about 60 hours. In 90 per cent oxygen they survived twice as long. In 80 per cent they did not die but were definitely ill when sacrificed after one week. In these environments there was a decline in the oxygen saturation of the arterial blood and a marked rise in hemoglobin concentration. Autopsies revealed extreme pulmonary congestion with interstitial edema and polymorphonuclear infiltration. In several dogs there were signs of right heart failure with

liver congestion. In some cases the pleural cavity contained fluid and fibrin. In all cases the spleen was intensely contracted, the intestines were empty and the stomach was distended. Control dogs maintained for a week in the chamber with 21 per cent oxygen were entirely normal.

In another series obstruction was produced just distal to a loop of the descending colon (4 dogs) or the terminal ileum (4 dogs) and gaseous distention developed spontaneously or was produced by introduction of air into the gut. These dogs were maintained in 90 to 100 per cent oxygen and 2 to 5 cc. samples of the intestinal gas were taken frequently for micro-analysis. Oxygen concentration in this gas increased for about 36 hours and then tended to decrease to very low levels before death coincident with large opposite changes in CO_2. These findings were consistent with the conclusion that oxygen poisoning begins to develop in dogs in 36 hours when the partial pressure of oxygen breathed is 700 mm. Hg or higher.

The excretion of sulfanilamide and endogenous urea by resting and stimulated submaxillary glands. ELIZABETH E. PAINTER and DOROTHY R. CHESS (introduced by M. I. Gregersen). *Department of Physiology, College of Physicians and Surgeons, Columbia University, New York City.*

Studies were made on 5 unanesthetized dogs with bilateral submaxillary fistulae. The "resting" secretion from each gland was 0.01 to 0.03 cc. per minute for 4 to 5 hours. The concentration of sulfanilamide in saliva was compared with that in whole blood and in plasma at levels of 4 to 5 mgm. per cent. Similar determinations were made for endogenous urea. When the concentrations are expressed per liter of water, the sulfanilamide in saliva is 20 per cent below that in whole blood and 5 per cent above that in plasma. The urea concentration was 8–9 per cent below that in blood.

When the secretion was increased to 0.2–0.4 cc. per minute by panting, the sulfanilamide in saliva was 30 per cent below that in whole blood. The concentration of urea was also 30 per cent lower. The water content of the saliva showed no appreciable change.

When the flow was increased to 3.0–5.0 cc. per minute by pilocarpine, the sulfanilamide and urea contents in saliva were 40 per cent below that in whole blood. It should be noted that increasing the rate of secretion has the reverse effect on the concentration of the electrolytes, sodium and chloride, in submaxillary saliva (Gregersen and Ingalls). With the pilocarpine stimulation a marked drop in the water content of the saliva is observed. This returns gradually to the control water level within one hour after stimulation and continues to rise through the second hour as the sulfanilamide content in the saliva rises.

Further data on the water content and sulfanilamide levels in saliva were obtained from two experiments in which adrenaline was injected over a period of 30 to 40 minutes at a rate of .0018 mgm. per kilo per minute. The submaxillary secretion was 0.1 to 0.2 cc. per minute. The water content of the saliva was increased and the sulfanilamide and urea showed the same concentration as that in whole blood. It would appear that the concentration of sulfanilamide and urea in saliva varies not only with the rate of flow, but is also dependent upon the water content.

Cinematographs of fetal behavior in bats. D. S. PANKRATZ (introduced by A. G. Mulder). *Department of Anatomy, University of Mississippi, Oxford.* (Motion picture demonstration.)

Cinematographs have been made of bat fetuses ranging from earliest motility to full term.

In fetuses 11–12 mm. there was ventral and lateral flexion of upper trunk accompanied by wing movement. The "flip reaction" was present in the wing, and the snout was sensitive to fine bristle stimulation. Fetuses 13–14 mm. showed head extension, opening and closing of mouth; and "flip reaction" and spontaneous movement in hindlimb. Bristle stimulation over rump of hindlimb caused hindlimb extension in 16–17 mm. fetuses. In 17 mm. and older fetuses, gasping and extensor movements occurred when the cord was ligated or when the mother was dying. Tongue movement occurred at 18 mm. In fetuses 19 mm. and older bristle stimulation of wing caused a typical scratch reflex in ipsilateral leg. Tail movements appeared at 21 mm. Fetuses of 24 mm. bit on bristle or grasped it with their feet. Fetuses 25 mm. and older grasped cloth and righted themselves. Movements became more definite and quicker with approaching parturition.

Desoxycorticosterone and corticosterone in the treatment of induced circulatory failure in the adrenalectomized dog. W. M. PARKINS, W. W. SWINGLE, J. W. REMINGTON (by invitation) and V. A. DRILL (by invitation). *Section of Physiology, Department of Biology, Princeton University, Princeton, N. J.*

Adrenalectomized dogs pretreated with intramuscular injections of desoxycorticosterone acetate (5 mgm. at 24, 18, 12 and 1 hour previously) are highly resistant to massive hemorrhage. Eighteen to 26 cc. of blood per kgm. body weight may be withdrawn from these unanesthetized animals, at the rate of 10 cc. per minute, without lowering the arterial pressure. Thirty to 35 cc. per kgm. are required to reduce the blood pressure to shock levels of 40 to 48 mm. Hg. The blood pressure returns to normal levels, with blood dilution, within 12 to 24 hours. The removal of only 5 to 16 cc. per kgm. from untreated dogs produces a fall in blood pressure to shock levels. Adequate compensation by vasoconstriction and significant dilution of the blood from extravascular fluid does not occur either during or following the hemorrhage. Death follows within a few hours unless cortical hormone is administered.

Although desoxycorticosterone acetate affords adequate protection against fatal circulatory collapse induced by muscle trauma, intraperitoneal glucose injections and epinephrine infusion, it gives no protection against shock resulting from intestinal stripping (Am. J. Physiol. **132:** 249, 1941). Recent experiments show that corticosterone will adequately protect against the circulatory failure resulting from this procedure. A relatively slight and transitory fall in blood pressure was observed with full recovery within 24 hours.

In our experience, dogs bilaterally adrenalectomized at one stage succumb within 10 to 24 hours unless large amounts of cortical extract are administered. Desoxycorticosterone pretreatment does not prevent the collapse following the surgical trauma when general anesthesia is used. When, however, in addition to the nembutal anesthesia, nerve plexuses

and sympathetic fibers in the vicinity of the glands are thoroughly bathed and infiltrated with novocaine, the dogs show only slight changes in blood pressure, remain active and eager for food, and survive indefinitely on daily maintenance doses of desoxycorticosterone.

Influence of anterior pituitary extracts on protein and carbohydrate metabolism. KARL E. PASCHKIS[1] (introduced by J. Earl Thomas). *Department of Physiology and Department of Medicine, Jefferson Medical College of Philadelphia.*

Extracts of the anterior pituitary gland enhance anabolism of protein. It has been generally assumed that this is a direct effect of the anterior pituitary gland. Mirsky on the basis of his experiments believes that protein anabolism is due to insulin, the output of which is stimulated by a pancreatotropic action of the pituitary extract. Decrease of blood sugar following the injection of pituitary extracts has been reported (Weinstein, Harrison and Long). These findings would tend to support Mirsky's theory.

The experiments on rats to be reported were carried out to determine:

a. Whether the effect on protein metabolism (as indicated by decrease in blood urea N) and the blood sugar lowering effect of an anterior pituitary extract (Antuitrin G., Parke Davis) were linked to one another. This should be expected if the effect on protein metabolism were secondary and due to pancreatic stimulation.

b. Whether either of the two actions is mediated through the adrenal.

c. Whether partial pancreatectomy would influence either of the two metabolic effects, as would be expected if they were mediated through the pancreas.

All experiments were carried out on rats fasted for about 16 hours.

Many of the intact animals responded with decrease of blood sugar. Decrease of urea N was present whether or not the blood sugar decreased. Also there is no parallelism of magnitude of decrease of the two constituents.

The drop in blood sugar following injection of the extract seems to be smaller in adrenalectomized rats but the percentage of animals showing a decrease is about the same as in the first group. The decrease of urea N did not differ from that observed in the intact animal.

In the partially pancreatectomized rats the decrease of blood urea was not different from that in the intact animal. Blood sugar decreased in some, in others there was a marked rise. In this group glucose excretion and nitrogen excretion were also studied. Large doses of Antuitrin G increased the glycosuria slightly but the nitrogen excretion was decreased.

The results indicate an independent mode of action of the pituitary extract on protein and carbohydrate metabolism.

Respiratory effects on the filling of the ventricles during vagal inhibition. MARY C. PATRAS (by invitation) and T. E. BOYD. *Department of Physiology and Pharmacology, Loyola University School of Medicine, Chicago, Ill.*

Ventricular volume was recorded by means of a cardiometer with closed

[1] J. Ewing Mears Fellow.

chest, the system being so arranged that external pressure on the ventricles follows the normal variations of intrathoracic pressure (see Boyd and Patras, accompanying paper). Diastole was prolonged by vagal stimulation.

During the expiratory pause the period of rapid filling is short and the curve of diastasis relatively flat. On the other hand, if the beginning of diastole coincides with the onset of inspiration, ventricular volume continues to increase as long as intrathoracic pressure is falling, the transition from rapid inflow to diastasis being delayed and much less clearly marked. The final volume reached, in a diastole of given duration, is much greater during inspiration than during the expiratory pause. The onset of either inspiration or expiration, if it appears during the period of diastasis, immediately changes the slope of the filling curve; with inspiration it becomes abruptly steeper, with expiration it is flattened.

If the recording tambour moves in outside air, so that the ventricles are left under atmospheric pressure, the above effects are reversed.

The reflex influence of the urinary bladder on the tonus and movements of the empty stomach of dogs. T. L. PATTERSON and L. E. DUNN (by invitation). *Department of Physiology, Wayne University College of Medicine, Detroit, Mich.*

States of abnormal distention in certain of the hollow viscera, manifest reflex changes in the normal activity of the heart, stomach and respiratory mechanisms. This work is primarily concerned with the reflex influence of increased intra-urinary bladder pressures on the gastric hunger motor mechanism and the pelvic nerve pathways through which these influences are mediated. Fistularized dogs were employed and gastric tonus and motility were recorded by the balloon manometer method, following fasts of 18 to 24 hours. After ascertaining the character of the tonus, motility and time relations of the different phases of the empty stomach, the dogs were provided with urinary bladder fistulae for the purpose of inserting a condom balloon to increase the intra-vesicular pressure at the same time of registration of gastric motility.

Later, neurectomy of either the "presacral," hypogastric or pelvic visceral nerves was performed through a mid-line laparotomy. Again, pressures were introduced in the urinary bladder and gastric motility recorded.

Minimal intra-urinary bladder pressures necessary to elicit gastric reflexes varied slightly from day to day, and if pressures were high subjective signs occurred in proportion to the degree of distention (yawning, restlessness and salivation). Pressures less than 32–35 mm. Hg do not produce any reflex effects on the tonus or motility of the empty stomach.

Pressures above 38 mm. Hg may reflexly cause one or more of the following: *a*, diminution in amplitude of the contractions; *b*, complete cessation of motility; *c*, loss of tonus; *d*, loss of tonus with inhibition; *e*, augmentation (slight) which rarely occurs.

After "presacral" or hypogastric nerve resection slightly greater pressures were required to elicit gastric reflexes. Section of the sensory components from the bladder produced no more dramatic results than section of the pelvic motor nerves in the sacral region. These results indicate that the chief nerve pathways concerned in gastro-urinary bladder reflexes are the "presacral."

The non-alcoholic 17-ketosteroids of neutral urinary extracts.[1] WILLIAM H. PEARLMAN (by invitation) and GREGORY PINCUS. *The Physiological Laboratories, Clark University, Worcester, Mass.*

A simple method has been devised by the authors whereby micro quantities of 17-ketosteroids may be partitioned into alcohols and non-alcohols. As little as 50 micrograms of dehydroisoandrosterone may be recovered in 90 per cent Zimmerman colorimetric titre by an adaptation of the macro method of succinic anhydride esterification. Similar quantitative recoveries were obtained when dehydroisoandrosterone and androsterone were added to urinary extracts.

The non-alcoholic 17-ketosteroid content of specimens of human urines was determined by this method. Procedures which are representative of those currently employed in the acid-hydrolysis of urine were compared. Significant quantities of such steroids were obtained regardless of the mildness of the procedure. Our finding that normal unhydrolysed male urine is very low in non-alcoholic 17-ketosteroid content indicates that such steroids exist chiefly in conjugated form or that they arise as artefacts from the hydroxy 17-ketosteroids during the course of hydrolysis.

The micro method of half-esterification with succinic anhydride permitted us to make a quantitative study of the effects of acid hydrolysis on hydroxy 17-ketosteroids. Considerable conversion into non-alcoholic material giving the Zimmerman color reaction occurred. In the case of dehydroisoandrosterone the non-alcoholic fraction also gave a red color in the Rosenheim test. These results are significant in connection with the recent isolation of $\Delta^{3,5}$ androstadienone-17 and androstenone-17 from human urines.

The method is useful in other phases of steroid investigations. In the case of the estrogens, micro quantities of non-ketonic estrogens, e.g., 100 micrograms estradiol, can be quantitatively separated from estrone since the phenolic hydroxyl group will not react with succinic anhydride under the conditions employed (Schwenk and Hildebrandt, U.S.P. 2, 046, 656, 1936).

Decerebellation in the rat. MORDANT E. PECK[2] (introduced by C. McC. Brooks). *Department of Physiology, the Johns Hopkins School of Medicine, Baltimore, Md.* (Motion picture demonstration.)

The entire cerebellum was removed from fifteen rats. Eight of these animals lived two months or longer. Their deficiencies, therefore, were considered to be permanent. Repeated studies justify the following observations:

1. Decerebellate rats maintain a broad base when walking and overcompensate when attempting to make any readjustments in their posture. During standing or sitting there is a hyperextension of the legs, an unnatural stiffness of posture and a general instability. The positive supporting reaction is exaggerated. When animals are suspended in the air their legs are abnormally extended and rigid.

2. Righting reactions are not noticeably deficient but the animals fail to make normal postural adjustments on landing. The result is an overcompensation which causes considerable rebound and oscillatory motions.

[1] Aided by grants from G. D. Searle & Co. and the Dazian Foundation for Medical Research.

[2] Henry Strong Denison Scholar.

3. Hopping reactions are delayed. The responses when they do occur are hypermetric, with the result that the legs are placed beyond the normal alignment.

4. Placing reactions likewise show hypermetria. In contrast to the normal, the decerebellate rat puts its paws beyond the edge of the test object then quickly withdraws them to the edge of the object.

5. The effect of these deficiencies on the animals is shown by their activity on a cross-bar. They are unable to maintain their footing, and hence to sit, walk or climb down normally.

Four to six months after operation the decerebellate animals were sacrificed and their brains studied. It was found that in no case had the vestibular nuclei or adjacent parts of the mesencephalon been injured.

The conclusion was drawn that the cerebellar syndrome in the rat consists of a marked decrease in activity, hypermetria in movements involving the peripheral musculature, and a generalized incoordination. It was thought that many of these deficiencies were different manifestations of the lack of synergistic action in the opposing muscle groups.

Basal metabolism in man after various doses of amphetamine sulphate. K. E. PENROD (introduced by Erma Smith). *Department of Physiology, Miami University, Oxford, O.*

After establishing the control levels for B. M. R. in each of 3 male subjects, (two obese and one of normal weight) the effect of amphetamine in 10, 20, 30 and 40 mgm. doses was determined by measuring the B. M. R. at 30 min. intervals for 3½ hrs. after each dose. Pulse rate, oral temperature and respiratory rate were recorded during each test.

All subjects showed slight increase in B. M. R. after the drug but significant elevations (+10 per cent to +14 per cent) occurred only in the subject whose weight is normal and in him only after the 30 and 40 mgm. doses. Pulse, temperature and respiration varied directly with B. M. R.

The influence of bile salts on active intestinal absorption of chloride. H. C. PETERS. *Department of Physiology, University of Tennessee, Memphis.*

Simultaneous active chloride absorption from a control solution of 0.5 isotonic sodium chloride and 0.5 isotonic sodium sulfate and a similar solution containing bile salts was studied in adjacent loops of lower ileum in anesthetized dogs. Sodium taurocholate and sodium glycocholate decreased chloride absorption in concentrations of 1.5 per cent. Sodium deoxycholate had this effect in a 0.2 per cent solution.

Cholate determinations, by the method of Reinhold and Wilson (J. Biol. Chem. **96:** 637, 1932), on the intestinal contents of acutely operated fat fed dogs, indicate that under physiological conditions concentrations which are toxic for the lower ileum do not exist in this region but do occur, however, in the duodenum. The lower ileum, where chloride is most rapidly absorbed against a concentration gradient, is apparently protected from the toxic action of bile salts by the diluting and absorbing function of the intestine above it.

Factors controlling the growth of rabbit blastocysts.[1] GREGORY PINCUS. *The Physiological Laboratories, Clark University, Worcester, Mass.*

[1] Aided by a grant from the National Research Council Committee for Problems of Sex.

Cleaved rabbit ova fail to enter the blastocyst stage when cultured *in vitro* in serum under anaerobic conditions. Blastocyst expansion is also inhibited by adding KCN to the culture medium (10^{-4} to 10^{-3} M). It is deduced, therefore, that energy for growth is derived from aerobic oxidative systems. A systematic investigation was, therefore, undertaken of the role of the various components of the usual cellular oxidative systems. The method employed involved the culture of 3-day rabbit eggs in blood serum under conditions involving a moderate blastocyst expansion. To the cultures were added various concentrations of theoretically active substrates and of enzyme poisons. The results may be summarized as follows: 1, glucose does not stimulate blastocyst growth; 2, the glucose concentration of the serum medium does not appreciably decrease over a period of four days' growth nor does it change in cultures containing ova inhibited from growing by poisons; 3, hexose phosphate fails to stimulate blastocyst growth; 4, phosphoglycerate is mildly growth-stimulating; 5, neither lactate, succinate, malate, fumarate nor glutamate proved growth-stimulating; 6, pyruvate is growth-stimulating in concentrations of 10^{-3} M to 10^{-2} M and growth-inhibiting in higher concentrations; 7, the growth-inhibiting effects of NaF are overcome by the simultaneous addition of pyruvate; 8, the growth-inhibiting effects of iodacetamide (10^{-4} to 10^{-3} M) are not overcome by the simultaneous addition of glucose, hexose phosphate, succinate, malate, fumarate or lactate, but are invariably overcome by the simultaneous addition of glutathione or cysteine; 9, cysteine and glutathione are invariably growth-stimulating; 10, partial growth inhibition is had with high concentrations of glyceraldehyde and malonate, the effects of glyceraldehyde are overcome by pyruvate. It is concluded that energy for growth is chiefly derived from the Meyerhof system of carbohydrate intermediaries, the role of sulfhydryl compounds being to maintain an iodoacetate-labile enzyme system. The relation of these findings to progesterone-controlled growth *in utero* will be indicated.

Respiratory responses from stimulation of the medulla of the cat. ROBERT F. PITTS. *Department of Physiology, New York University College of Medicine, New York City*

Two years ago at the society meetings at Toronto, evidence was presented that the medullary respiratory center of the cat is composed of two divisions, both confined to the reticular formation of the caudal part of the medulla. A ventral inspiratory center overlies the cephalic four-fifths of the inferior olivary nucleus while more dorsally and slightly more cephalically is situated an inspiratory-inhibitory or "expiratory" center (Pitts, Magoun and Ranson). Recently the method of exploratory stimulation upon which the above conclusions were based has been criticized by Brookhart (Am. J. Physiol. **129:** 709, 1940), along three lines: 1, excessive spread of stimulus; 2, with such stimulus spread a functionally meaningless differentiation of the respiratory center into antagonistic divisions; 3, criteria for differentiation of inspiratory and expiratory centers which are too all inclusive.

The degree of stimulus spread has been assessed by stimulation of the descending root of the trigeminal nerve in the medulla and recording the impulses conducted antidromically over the frontal branch of the ophthalmic division of the fifth nerve. Utilizing this method it has been found that the stimulus diminishes rapidly in intensity over a distance of 0.5 mm. fron the electrode tips.

An evaluation of the method of exploratory stimulation for the localization of the descending trigeminal root in its course through the medulla has demonstrated the adequacy of the method under the conditions of our experiments, namely bipolar needle electrodes, repetitive condenser discharges at a time constant of 0.1 millisecond and an intensity of 8 volts. A repetition of the localization of the respiratory center has yielded results in agreement with those originally reported by us and summarized in the first paragraph.

The criteria for localization of inspiratory and expiratory centers; namely, maintained deep inspiration or maintained expiration have been examined in terms of the activity of single fibers of the phrenic nerve. It has been found that stimulation of the ventral inspiratory center converts the bursts of repetitive impulses of phrenic fibers into a maintained repetitive discharge at a greater than normal frequency. Stimulation of the dorsal "expiratory" region causes a complete cessation of activity in phrenic fibers.

Edema of the pancreas. H. L. POPPER (by invitation) and H. NECHELES. *Department of Gastro-Intestinal Research, Michael Reese Hospital, Chicago, Ill.*

Experimental production of edema of the pancreatic gland was attempted in dogs. It is known that mere ligation of the ducts of the pancreas is not followed by edema. On the other hand, injection into the ducts of various substances and subsequent ligation of the duct is followed by the formation of edema within a few minutes. In our experiments, dogs were anesthetized, and either the main or the main and accessory pancreatic ducts ligated. Simple ligation was not followed by edema within sixty minutes. When a solution of 40 mgm. of secretin was administered intravenously, a distinct edema spread over the entire pancreas within 3 to 5 minutes. In a number of control experiments secretin was injected before ligation of the ducts, without effect. Injection of mecholyl and eserine or of pilocarpine was not followed by pancreatic edema in animals with the pancreatic ducts ligated. This is explained by the inferior secretagogue effect of the latter drugs as compared to secretin. Injection of 0.05 cc. of dogs' duodenal juice into and ligation of the pancreatic duct did not lead to edema of the pancreas. Subsequent subcutaneous injection of pilocarpine was followed by distinct edema of the pancreas.

These experiments show that the formation of edema of the pancreas depends on the volume of fluid accumulating in the gland. From previous experimental experience we assume that these retained secretions are activated within the gland, because the edema fluid aspirated by puncture of the capsule contains highly active and rather concentrated enzymes. The bearing of the above results on edema of the pancreas in the human seems evident. Obstruction of a pancreatic duct by gallstones or by a spasm of short duration may lead to edema of the gland if the latter is stimulated by secretin or other mechanisms.

An in vitro study of lower vertebrate endocrine organs. ETHEL G. PORIS, ALBERT S. GORDON, IRVING LEVENSTEIN and HARRY A. CHARIPPER (introduced by Robert Gaunt). *Department of Biology, Washington Square College of Arts and Science, New York University, New York City.* (Read by title.)

The techniques of Parker and Carrel have been employed in studying, in tissue culture, the behavior of endocrine organs of Anolis carolinensis, Xenopus laevis, Necturus maculosus and Rana catesbiana and pipiens tadpoles. 2 cc. of the medium which consisted generally of two parts adult guinea pig or rabbit serum, one part amphibian Tyrode's, and phenol red (0.005 per cent) were introduced into 75 cc. capacity Carrel flasks. About 5–10 whole organs or organ fragments were then placed into the medium. A gas mixture consisting of 92 per cent oxygen and 8 per cent carbon dioxide was led once daily into the flasks. The pH was kept between 7.2 and 7.5, and the temperature at 25°C. Among the organs successfully maintained, for 4 or 5 day periods, were the pituitary, thyroid, testis and ovary of Anolis, the pituitary and testis of Necturus, the testis and ovary of Xenopus, and the pituitary of Rana tadpoles. These results were, on the whole, more satisfactory than those obtained, under similar conditions, with mammalian endocrine organs (Levenstein, Gordon and Charipper. Proc. Soc. Exper. Biol. and Med., in press). Organs placed into amphibian Ringer's, amphibian Tyrode's or in media containing less than 50 per cent serum revealed degeneration after 4–5 days.

Further experiments were conducted on the testis of Anolis. After five days of cultivation, the tubules showed a marked increase in the numbers of spermatogonia and spermatocytes as compared with non-cultured control organs. Introduction of 5 or 10 R.U. pregnant mare serum hormone caused a stimulation of spermatogenesis but had no apparent effect on the interstitial tissue of cultivated immature Anolis testis. The development of Anolis testis was unaffected by estrone added to the flasks in 1, 5, or 20 I.U. quantities. Further experiments designed to test the effects of different hormones on various lower vertebrate endocrine organs are contemplated.

Summation of spinal reflex action by intra-arterial injection of potassium chloride. EUGENE L. PORTER, HUGH ARNOLD (by invitation) and W. H. GRANGER (by invitation). *Department of Physiology, Medical School of the University of Texas, Galveston.*

Stimulation of the posterior tibial nerve through a liquid electrode of special form gives relatively uniform reflex contractions of Tibialis anticus muscle. These are recorded when stimulation is just above threshold. Stimuli are given rhythmically about once in four seconds.

A bent hypodermic needle is inserted in the femoral artery. It is supported and remains there throughout the experiment. A very slow (1 cc./min.) steady stream of Ringer Solution from a burette flows through the needle into the artery in order to keep the needle open. A three way stop-cock is included in the tubing from burette to needle.

A dilute solution of Ringer + KCl is now injected into the artery through the stop-cock. The contractions of Tibialis anticus increase in height. This effect occurs only when the sensory nerve is stimulated. The increased height is therefore a summation effect, i.e., "sensation" from artery in added to "sensation" from the stimulated nerve. This summation has been observed when the amount of KCl is only double the normal amount in Ringer's Solution. This makes the concentration around 0.1 per cent KCl. This is still further diluted by the blood flowing past the inserted hypodermic needle.

Similar injections of Ringer Solution or Ringer + $CaCl_2$ have no effect or a very slight transitory one.

The amount of KCl causing "sensation" as indicated by summation in these experiments is approximately one-third that found by Moore (Am. J. Physiol. **107:** 594, 1934) when the criterion of sensation was actual movement and vocalization started by the injection alone in the lightly anaesthetized animal.

Phasic inflow patterns in femoral and carotid arteries.[1] W. H. PRITCHARD (by invitation) and D. E. GREGG. *Department of Medicine, Western Reserve University, Cleveland, O.* (Read by title.)

Comparisons have been made of phasic inflow curves in femoral and carotid arteries of dogs by use of the orifice meter (Gregg and Green. Am. J. Physiol. **130:** 114, 1940). These were recorded with their respective pressure pulses.

In the femoral artery, with the onset of systolic pressure rise, the flow accelerates abruptly from zero, reaching a maximal velocity coincident with maximal systolic pressure. The flow then decelerates rapidly to the first incisura and then more gradually to the diastolic dip where a small backflow is usually present. Subsequently, the flow augments slowly, reaching zero or slightly positive values toward the end of diastole.

The contour of the carotid curves resembles closely the carotid pressure pulse and at no time does flow velocity reach zero.

The factors causing these differences are being studied.

Blood flow in the right coronary artery.[1] W. H. PRITCHARD (by invitation), D. E. GREGG, A. ROTTA (by invitation) and J. DINGLE KENT (by invitation). *Department of Medicine, Western Reserve University, Cleveland, O.*

The phasic inflow at the right coronary orifice has been determined in anesthetized dogs using the orifice plate meter (Gregg and Green. Am. J. Physiol. **130:** 114, 1940). This has been compared with the differential pressure curve simultaneously determined from direct measurements of aortic and peripheral coronary pressures.

In good hearts the orifice inflow curve decreases mildly during isometric contraction, rises rapidly during ejection with the aortic pressure, reaches a peak during the middle of the rise of aortic pressure, and then abruptly declines during the latter part of systole. Following the incisura, there may be a momentary acceleration of flow or a progressive deceleration of flow throughout diastole as the aortic blood pressure falls. This flow curve is patterned after the central coronary pulse and at no point in systole (or diastole) does the rate of flow approach zero as it so often does in the left coronary artery.

The orifice curve is similar to the differential pressure curve in timing and contour but is slightly greater during mid-systole and most of diastole. These small differences are explained; in part by the errors involved in the experimental determination and reconstruction of the two curves; in part by the compressor action of ventricular muscle on coronary vessels and the volume elastic effects produced by the aortic pressure.

Further comparisons are being made between total inflow and differential pressure curves obtained under various dynamic conditions such as

[1] Aided by a grant from the Commonwealth Fund.

elevation of right ventricular and aortic pressure, cardiac venous occlusion and increased venous return.

Effects of ions upon isolated nerve centers. C. LADD PROSSER. *University of Illinois, Urbana.*

The spontaneous activity of nerve centers can be altered with respect to *a*, the number of active units; *b*, frequency of discharge of individual units; *c*, integration of units into summed rhythmic waves, and *d*, frequency of the integrated waves. Various kinds of nerve centers are affected differently by ionic variation of their medium. When crayfish abdominal ganglia are bathed with a saline solution low in potassium the gross spontaneous discharge increases, due almost entirely to a great increase in number of active neurones. Solutions high in potassium have an opposite action. Bivalent cations have very little effect. Monovalent cations are effective to reduce the number of active units in the following order: $K > Cs > Rb > NH_4 \gg Na > Li$. The anions Cl, Br, NO_3, acetate, are without effect, indicating anion impermeability. Thiocyanate, and to a less extent iodide, are stimulating in action. The calcium-precipitating anions, citrate, tartrate, and sulphate, have a stimulating effect. A close correlation between spontaneity and resting potential is indicated.

In contrast to the crayfish abdominal ganglia the isolated cardiac ganglion of Limulus responds to high potassium with an increase in frequency of discharge in single neurones and an increase in the gross frequency of the cardiac rhythm. Low potassium acts oppositely. Also unlike the crayfish abdominal ganglia the potassium effects on the Limulus heart are readily antagonized by calcium. The above results taken together with data on other systems (e.g., frog brain by Libet and Gerard, J. Neurol. **2:** 153, 1939), indicate that a spontaneously active nerve center may be influenced in either or both of two quite separate ways by potassium. The number of active units (amplitude) or the frequency of discharge may be affected.

The influence of fats and related substances on the motor activity of the pyloric sphincter region and on the process of gastric evacuation. J. P. QUIGLEY, I. MESCHAN (by invitation), J. M. WERLE (by invitation), E. W. LIGON, JR. (by invitation), M. R. READ (by invitation) and K. H. RADZOW (by invitation). *Department of Physiology, Western Reserve University Medical School, Cleveland, O.*

Investigating in fasting trained dogs the mechanism by which fats retard gastric evacuation we have learned by the triple balloon technic method of Meschan and Quigley (Am. J. Physiol. **121:** 354, 1938) (a balloon in the antrum, sphincter and bulb) that cream introduced into the duodenum causes motor inhibition of these three regions. Thus gastric evacuation is retarded because of the decreased pumping action of the antrum and in spite of the decreased resistance offered by the sphincter and bulb.

Employing fluoroscopic observations and optical registration of antral and bulbar pressures, Werle, Brody, Ligon, Read and Quigley (Am. J. Physiol. **131:** 606, 1941) obtained evidence that gastric evacuation results from a pressure gradient from antrum to bulb combined with propulsive antral peristalsis. We have employed the same methods and found that in fasting dogs, cream introduced into the duodenum preferentially de-

pressed antral basal pressure so the antral-bulbar pressure gradient was reduced or even reversed. The degree and duration of the effect was related to the volume of cream administered. After introducing 300 cc. of a corn meal mush—$BaSO_4$ mixture into the stomach, cream in duodenum produced the effects noted in fasting animals, but the magnitude of action was decreased. Larger volumes of cream were required to depress antral basal pressure and the antral-bulbar gradient was less frequently reversed. Retarded gastric evacuation was closely related to and primarily dependent upon suppression of antral propulsive peristalsis.

Cream in the duodenum modified pressures and motility of the sphincter region less in the fed than in the fasting animal. This was due in part to the exaggerated antral pumping activity incident to gastric distention in the fed animal, for a balloon containing 500 cc. of air placed in the stomach of a fasting animal produced results comparable to those noted in fed animals. Dilution and flushing out of the cream by the chyme discharged into the duodenum also decreased the effect of cream in the fed animal. Prevention of this effect by duodenal drainage increased the effectiveness of cream in the fed animal.

Further studies of the influence of certain steroid hormones on the diffusion of sodium and chloride into the peritoneal space. A. E. RAKOFF (by invitation) and A. CANTAROW. *Departments of Obstetrics and Medicine, Jefferson Medical College and Laboratory of Biochemistry, Jefferson Hospital, Philadelphia, Pa.*

It has been demonstrated that administration of desoxycorticosterone acetate or progesterone to dogs results in a marked increase in the rate of entrance of Na and Cl into 5.5 per cent dextrose solution introduced into the peritoneal space. The same effect upon diffusion of Cl was observed following oral administration of desoxycorticosterone acetate and intramuscular injection of estradiol, diethylstilbestrol and testosterone proprionate. The increased rate of diffusion of these ions was most marked during the first 30 minutes of the experiment. The effect of the hormones was usually most pronounced 24–48 hours after their administration, but could be demonstrated for as long as 96 hours.

This phenomenon could not be demonstrated in rabbits following administration of desoxycorticosterone acetate, progesterone or diethylstilbestrol. Pregnancy did not materially influence the rate of diffusion of Cl into the peritoneal fluid in rabbits.

The experiments in dogs support the hypothesis that the effect of these hormones in diminishing renal elimination of Na and Cl may be a reflection of an action exerted upon membranes in general. The failure to demonstrate a similar effect in rabbits indicates the presence of an important species difference in this regard.

The effect of insulin on the metabolism of vitamin C.[1] ELAINE P. RALLI and SOL SHERRY (by invitation). *The Laboratories of the Department of Medicine, New York University College of Medicine, New York City.*

It has been reported previously (Ralli and Sherry) that on identical diets totally devoid of vitamin C, depancreatized dogs excreted less vitamin C than normal dogs. This was found to be related to the administration of

[1] This research was aided by a grant from the Josiah Macy, Jr. Foundation.

insulin which the diabetic dogs received to control the glycosuria. When insulin was injected into normal and diabetic dogs and into diabetic patients, and the plasma levels of vitamin C were determined at short intervals, it was found in each instance that a fall occurred in the level of vitamin C of the plasma. The rapidity of this depended on whether the insulin was injected intravenously, intramuscularly or subcutaneously. The decrease in the plasma vitamin C was accompanied by a decreased urinary excretion of the vitamin. It has now been observed that insulin has no effect on vitamin C in vitro; that the fall in the plasma level of vitamin C in the dog, an animal which ordinarily synthesizes this vitamin, is not due to any interference in the synthesis is of vitamin C and thirdly that insulin does not increase the utilization of vitamin C. Apparently the effect of injected insulin in both dogs and humans is to cause a redistribution of vitamin C between the blood plasma and the tissues. This is borne out by the fact that as the plasma level of vitamin C falls there is a concomitant rise in the level of vitamin C in the white cell platelet layer. Only about 30 per cent of the amount of vitamin C disappearing from the plasma could be accounted for by the rise in the white blood cell platelet layer. Analysis of the vitamin C in the red blood cells at the time the plasma level had fallen did not show any increase in vitamin C. Other observers, (Crandon et al. and Butler and Cushman) have shown that the white cell concentration of vitamin C reflects most closely the concentration of vitamin C in the body tissues. It seems probable to us, that the 70 per cent of vitamin C not accounted for passes into the other body tissues. That the plasma vitamin C was shifted and not lost from the body, is suggested by the fact that in experiments carried out for 12 hour periods it was found that after the fall in the plasma level and urinary excretion of vitamin C following insulin, there was later a rise in the plasma level above the original fasting level and a little later this returned to the original normal level. The urinary excretion reflected the changes in the plasma. This suggests a release of the vitamin from the tissues after the effect of insulin has worn off.

The relations of the absolutely refractory period, relatively refractory period and tension in isolated muscle fibers of the frog. ROBERT W. RAMSEY and SIBYL F. STREET (by invitation). *Department of Physiology, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* (Read by title.)

Isolated uninjured muscle fibers of the frog were stimulated through a non-polarizable unipolar pore electrode by either direct current rectangular shocks or by condenser discharges with a duration (respectively, time constant) or 0.3 msec. corresponding to a 3 to 4 x rheobasic shock. The relatively refractory period was explored with test shocks, threshold at each point, up to a maximum of 25 rheobases. Isometric tension was recorded by photographing the magnified deflections of a quartz cantilever.

The results and conclusions were as follows:

1. Both the propagated and local excitatory process have an absolutely refractory period varying from 12.5 to 5.00 sigma over a range of temperature from 8.5° to 14°C.

2. Modifications of twitch tension brought about by change of temperature or by the effect of previous activity are always accompanied by corresponding charges in the duration of the absolutely refractory period. The correspondence is not always linear.

3. If at one temperature, the initial twitch tension to tetanus tension ratios for different fibers are plotted against the durations of their absolutely refractory periods an approximately linear relation results.

4. At low temperature the twitch tension approaches the tetanus tension in magnitude. Under these conditions an increase in the absolutely refractory period is accompanied by an increased duration of the twitch rather than increased tension.

5. The added increment of tension due to a second shock applied during the relatively refractory period of the first is not constant but increases to a maximum with increase in the interval between the two shocks (provided the interval is not so long that incomplete summation results).

6. The rate of development of tension of the summated response from two shocks is greater the earlier the second shock falls in the relatively refractory period of the first.

The relation of serum proteins to the effect of horse serum and serum ultrafiltrate on tissue metabolism. DAVID RAPPORT, ATTILIO CANZANELLI, GERTRUDE ROGERS (by invitation) and CLEMENT S. DWYER (by invitation). *Department of Physiology, Tufts College Medical School, Boston, Mass.*

In the following observations, the O_2 consumption of guinea pig tissue slices, determined in the Warburg apparatus, is expressed in relation to that found when the tissue is respiring in a medium of 0.15 M NaCl buffered with phosphate.

a. *Liver.* In horse serum the O_2 consumption is increased to a maximum of 125 per cent (the effect being roughly a straight line function of the proportion of serum in the medium). Horse serum proteins also increase the QO_2, up to a maximum of about 45 per cent for serum globulin, and of about 35 per cent for serum albumen. However, the protein-free ultrafiltrate of serum raises the O_2 uptake to the same extent as serum itself. The effects of serum globulin are not summated with either the effects of serum or of its ultrafiltrate. "Pasteurization" or boiling does not alter the effect of the ultrafiltrate. The presence or absence of glucose alters none of the above effects.

b. *Brain.* The effects of serum protein in $NaCl/PO_4$ are influenced by the presence or absence of glucose in the medium. When glucose is present both serum globulin and serum albumen depress the brain O_2 uptake; when it is absent they either have no effect or increase the O_2. Both in serum and its ultrafiltrate (with or without added glucose) the brain O_2 uptake is lower than in $NaCl/PO_4$ with glucose added, though much higher than in $NaCl/PO_4$ without glucose.

c. *Kidney.* Serum globulin raises the O_2 consumption of kidney as much as 36 per cent, (average 16 per cent), but in some cases there is no stimulation at all. The presence of glucose in the medium abolishes the globulin effect, when obtained. Serum albumen is without effect. Both serum and its ultrafiltrate, on the other hand, increase the O_2 consumption markedly (60 per cent) in the absence of glucose from the medium; less so when glucose is present in the latter. Glucose, which raises the O_2 uptake of kidney in $NaCl/PO_4$ (to a maximum of about 25 per cent), has no effect when the tissue is respiring in either serum or ultrafiltrate.

Interaction of adjacent intraspinal mammalian axons. BIRDSEY RENSHAW and PER OLOF THERMAN (introduced by Herbert S. Gasser). *Laboratories of The Rockefeller Institute for Medical Research, New York City.*

The excitation of axons by the activity of adjacent axons, first demonstrated by Hering (1882) in frog nerve, has been confirmed for intraspinal mammalian axons (cat). The ascending branches of dorsal root fibers lie in the ipsilateral dorsal column. After transection of the dorsal column at a level cephalad to the entry of a stimulated dorsal root, impulses in the ascending branches of the active fibers directly excite adjacent axons. The impulses in the secondary axons then travel antidromically (caudally) and emerge as a centrifugal discharge in dorsal root fibers adjacent to those which carried the centripetal volley. The secondary impulses are initiated before post-synaptic spinal neurons become active; in fact, the centrifugal volley arrives at the recording electrodes only 0.1 to 0.3 msec. later than it would if the primary (conditioning) impulses had travelled in an uninterrupted fiber path from the stimulating cathode cephalad to the region of the cut and back to the recording electrodes.

The number of secondary fibers which are excited by a given conditioning volley decreases progressively with the time elapsed after transection of the dorsal column. After the primary volley has ceased to initiate secondary impulses, it continues to induce subliminal excitability changes in tested secondary axons. The subliminal changes may be measured by changes in the number of tested axons that are stimulated by a submaximal shock applied to the dorsal column. As Blair and Erlanger have shown for frog nerve, at loci some distance from an injured region the excitability of the tested axons is *decreased* as the primary impulses pass in adjacent axons. Only near the transection (within 5 to 7 mm.) are excitability *increases* induced by the primary impulses. These increases are largest 2 ± mm. caudad to the transection, and they occur during the period of negativity which is produced by the conditioning impulses.

It is, therefore, inferred that the locus of the initiation of secondary impulses is within a few millimeters of the transection. The secondary impulses arise at approximately the time of arrival at this locus of the negative peak of the spike potential in the primary axons (350 words).

Sensitivity of the smallest blood vessels in normal human skin: responses to graded mechanical stimulation in normal men.[1] S. R. M. REYNOLDS, J. DI PALMA (by invitation) and F. I. FOSTER (by invitation). *Department of Physiology and Pharmacology, Long Island College of Medicine, Brooklyn, N. Y.*

By application of selected weights at appropriate rates of speed, it is shown to be possible to elicit threshold responses of the smallest blood vessels of the skin. Such a response consists of an uneven, incomplete line of red, resulting from vasodilatation, along the line of the stroker surrounded by a restricted region of pallor (vasoconstriction); a subthreshold response is characterized by the vasoconstriction alone, superthreshold responses, by a continuous, well-defined line of hyperemia along the line of the stroker surrounded by a pale area. A machine is described by which

[1] Supported by grants from the Josiah Macy, Jr. Foundation and the Committee on Endocrinology, National Research Council.

these effects are easily obtainable at will. It is found that a typical time-intensity (strength-duration) curve is obtainable. From this a coefficient of excitability is calculated. By experiment, the effect on sensitivity of the smallest blood vessels of partial and complete circulatory occlusion is measured. The fact is established that pronounced changes in sensitiviy occur with anoxemia (a decrease in sensitivity) and hypercapnea (an increase in sensitivity) before any important changes in cardiac or respiratory activity take place. With a change in surface temperature, about a two per cent change in threshold occurs per degree Fahrenheit. By use of this technic, the effect of age, sex, climate, disease and pharmacologic or therapeutic procedures on the excitability of the smallest blood vessels can be estimated.

The action of quinidine sulfate on the respiration of rat tissue slices. JAMES C. RICE and FRED G. BRAZDA (introduced by Clyde Brooks). *Departments of Pharmacology and Experimental Therapeutics and Biochemistry, Louisiana State University School of Medicine, New Orleans.*

The study herein reported is one of a series of related studies employing the concept of concentration-action curves in the investigation of the fundamental mode of action of drugs on cells and on enzyme systems. In this case depression of oxygen utilization, as determined by the Warburg technique, serves as the measure of action of the protoplasmic poison quinidine.

In the presence of 0.2 per cent glucose in buffered saline (pH 7.4) the concentration-action curve relating the per cent of depression of oxygen uptake of rat kidney slices to concentration of the drug in the range 2×10^{-4} to 2×10^{-3} molar is gently convex with respect to the axis of the abscissa. The slope of the curve is steeper with low concentrations and becomes progressively less steep through the tenfold increase in concentration of the drug. The range of depression for the concentrations studied varies from about 16 per cent to 40 per cent.

The form of the curve suggests reversible changes, and not a preponderance of effects predicated on variability of the cell populations represented in the slices.

Studies with liver slices which are, as yet, not completed, suggest a similar curve with a much steeper slope in the low concentration range. The slope becomes less steep with increase in concentration, but remains steeper than that of kidney slices.

A simple fluorescence technic for demonstrating acid-fast bacteria. OSCAR W. RICHARDS. *Research Department, Spencer Lens Co., Buffalo, N. Y.* (Demonstration.)

Filters, an aluminized mirror, and an intense, concentrated filament lamp operating at a low voltage have been devised for attachment to a monocular microscope to make possible the use of the fluorescence technic for the rapid identification of acid-fast bacteria. Under the ultraviolet radiation, acid-fast bacteria that have been stained by carbol auramin and the background decolorized by a special acid-alcohol appear as bright yellow bodies against an almost black background. The great contrast between the self-luminous bacteria and the dark background permits the organisms to out stand brilliantly at a magnification of 400 diameters (8 mm. objective

and 20 × ocular or 4 mm. objective and 10 × ocular). The large visual field obtaining under these circumstances greatly facilitates the study of smears of sputum and other materials. (Staining is done at room temperature and no counterstain is used.) Comparative studies of the fluorescence technic and the Ziehl-Neelsen method have demonstrated that acid-fast bacteria can be detected in direct smears of sputum, spinal fluid, and purulent exudate by the fluorescent method when they can not be detected by the Ziehl-Neelsen technic. The fluorescence technic has been found to be slightly more sensitive on direct smear than the Ziehl-Neelsen method on concentrated sputa samples. More bacteria are seen due to a firmer combination of the auramin than the fuchsin with the mycolic acid of the tuberculosis bacteria.

Intracisternal administration of picrotoxin. R. KOHN RICHARDS, CLYDE GRIMES (by invitation) and ALBERT SMITH (by invitation). *Abbott Laboratories, North Chicago, Ill.* (Read by title.)

On account of the latent period following even an intravenous injection of Picrotoxin, the question of possible intracisternal injection in desperate cases of barbiturate poisoning is raised. Rice and Isenberger (J. Pharmacol. **59:** 43, 1937) found that Picrotoxin intracisternally shortens the respiratory paralysis following intracisternal injection of Amytal in dogs. No data on intracisternal toxicity and therapeutic range are available. Rabbits were injected intracisternally with an 0.3 per cent solution of Picrotoxin, the total volume not exceeding 0.5 cc. All animals on which a possible injury to the medulla or bleeding occurred were discarded. Three of 7 animals died on 7, 8 of 14 on 10, and 8 of 10 on 15 gamma per kgm. Thus, the LD50 is approximately 10 gamma/kgm. against 1.25 mgm./kgm. by intravenous administration. There is a latent period of only one minute until predominantly tonic convulsions start which later change into the typical Picrotoxin convulsions. If the animals are injected with 25 mg./kg. pentobarbital (Nembutal) intravenously 10 minutes before the intracisternal administration of Picrotoxin, the LD50 for the latter is raised to about 150 gamma/kgm. This is a wider margin than can be obtained by the intravenous administration of Picrotoxin against the same dose of Nembutal. Similarly, the lethal dose of Nembutal can be somewhat increased, by properly antagonizing it by intracisternal Picrotoxin administration. While improvement of respiration and twitching follows the intracisternal Picrotoxin injection of rabbits in Nembutal anesthesia, no real analeptic effect as evidenced by shortening of the sleep can be produced by this method. This is probably due to failure of adequate amounts of Picrotoxin reaching the higher centers. It is thought that in desperate clinical cases a cautious intracisternal administration of Picrotoxin may be tried and followed by the regular intravenous treatment.

Studies on the toxic effects of stilbestrol with special reference to growth retardation. R. KOHN RICHARDS and KENNETH KUETER (by invitation). *Abbott Laboratories, North Chicago, Ill.*

The question of toxic side actions of stilbestrol in humans and animals is still debated by various authors. Studies on white rats of different ages at the Abbott Laboratories, in which stilbestrol in oil was fed in doses up to 10 mgm. daily up to 60 days, failed to produce liver necrosis. The organ changes observed were the same as those which occurred with pro-

longed feeding of plain oil. Occasional bleeding occurred in the adrenals of old females. The retardation of growth observed with chronic administration of stilbestrol raised the question of whether this was due to a direct toxic action or an inhibition of the secretion of pituitary growth hormone. Male and female rats weighing about 100–130 grams were fed with 2 mg. of stilbestrol daily until a definite drop of weight occurred. While this treatment was continued, 0.5 cc. of pituitary growth hormone of the Wilson Laboratories was injected daily for 10 days. This resulted in a return to the normal growth rate for the duration of the injection period. Following cessation of these injections but with stilbestrol administration continued, the weight dropped again. Other experiments seem to indicate, however, that the growth depressing effect of stilbestrol becomes less on prolonged administration of this substance. It is concluded that the depression of growth is due to an inhibition of the growth hormone secretion by the anterior lobe.

Calcium appetite of rats used to bioassay substances affecting blood calcium. CURT P. RICHTER and JOHN R. BIRMINGHAM (by invitation). *Psychobiological Laboratory, Phipps Psychiatric Clinic, Johns Hopkins Hospital, Baltimore, Md.*

Previous experiments showed that parathyroidectomized rats have a greatly increased appetite for calcium solutions. Some of the rats drank 10 to 15 times as much of these solutions after parathyroidectomy as before. It has been found that the calcium appetite is a very sensitive indicator of the activity of substances which affect blood calcium. Dihydrotachysterol (A T 10) given in the food reduced the calcium appetite within 24 hours. Doses as small as 30 gamma per day sufficed to decrease the calcium appetite to its normal level. With this method we determined and compared the minimum amounts necessary to reduce the calcium lactate appetite to its normal level for the following substances: irradiated ergosterol, irradiated cholesterol, crystalline vitamin D_2 and D_3, cod liver oil, and parathormone.

The variability of human stance patterns. KATHRYN RIDDLE (by invitation) and F. A. HELLEBRANDT. *University of Wisconsin, Madison.* (Read by title.)

The center of gravity of the body shifts incessantly during standing. When the stance is relaxed and comfortable the displacement may approach in magnitude the antero-posterior diameter of the middle third of the supporting base. The object of this experiment was to determine the influence of postural sway on the validity of a stance measure dependent upon instantaneous photography. Approximately 1000 observations were made on 7 young adult women. The stance was scored by the Wellesley method which statistically weights the antero-posterior curvature of the spine, head poise, chest position and body lean.

The subect stood for five minutes, during which photographs were taken at 5 second intervals. The best stance attainable by strong voluntary effort was compared with a natural, comfortable posture. As might be predicted *a priori* the maximal variations in score occurred in subjects showing the greatest involuntary shift in the center of gravity during standing. They ranged from 5 to 17 or a letter grade of A− to D. Sig-

nificant variations in alignment may occur within 5 seconds of time. When a relaxed stance is habitually assumed, voluntary correction cannot be maintained and the alignment deteriorates progressively, commencing within the first half minute of standing. When the equilibrium is good and postural sway is minimal, relaxed and corrected postures are maintained without sign of deterioration, and insignificant changes in stance score occur, being limited to the extremes of one letter grade.

Some effects of eschatin and acetyl choline on the contractions of striated muscle in the cat. SARAH R. RIEDMAN (by invitation) and HELEN C. COOMBS. *Department of Hygiene, Brooklyn College, and the Department of Physiology, New York Medical College, Flower and Fifth Avenue Hospitals, New York City.* (Read by title.)

In a previous paper, the effects of changes in the concentration of calcium were discussed in relation to the metabolic processes in muscle, as indicated by its working power and the onset and degree of contracture.

In this series of experiments, following the procedure outlined in the earlier paper,[1] the effects of stimulating the rectus abdominis in the cat were studied under the influence of eschatin and acetyl choline. (Nembutal was used as the anaesthetic.) While still in vascular continuity with the general circulation, excitation of the rectus abdominis was done with a constant strength of stimulus at the rate of one per second. The contractions were recorded continuously before and after intravenous injection of 2 cc. of eschatin (Parke Davis). The work performed was computed and the onset of contracture observed.

The tabulation of a typical experiment follows:

EXPERIMENT	WORK DONE (GM. CM.)		PER CENT VARIATION
	Control rectus	Rectus after eschatin	
20	3477	7060	Plus 103

In a series of ten experiments, the increase in working power of the muscle following administration of eschatin ranged between 30 per cent and 164 per cent. Four control experiments show that the right and left rectus perform practically the same amount of work and may be used interchangeably. For example, in experiment 13, the right rectus performed 6068 gm. cm. and the left rectus 5885 gm. cm. of work—a variation of approximately 3 per cent. The shape of the curve obtained seems to be characteristic of the animal.

The effect of eschatin on the onset of contracture appears with the same consistency as the effect upon the work performed—namely, that contracture in the experimental muscle either totally disappears or is markedly reduced, and the onset of fatigue delayed, in comparison with the control muscle.

When acetyl choline is administered in the same manner, possibly due to its rapid dissociation, the effects are less marked than with eschatin, although there is a definite increase of the total work performed (from 4 to 29 per cent) (six experiments). Moreover, the onset of contracture

[1] H. C. Coombs, F. H. Pike, and D. S. Searle. Endocrinology **19**: 421, 1935.

appears early in the curve and is much exaggerated; with eschatin, contracture is almost absent.

A more detailed report is in preparation.

Quantum efficiencies of photosynthesis. FOSTER RIEKE (introduced by E. M. K. Geiling). *University of Chicago, Chicago, Ill.*

Quantum efficiency measurements have been made on thin suspensions of unicellular green algae. The advantage in using thin suspensions (approximately 30 per cent absorption) is that one avoids excessively large corrections for respiration. The absorption of light by such suspensions, which scatter as well as absorb light, can be measured accurately and conveniently in an integrating sphere. Efficiencies have been measured both for the usual aerobic photosynthetic process in which CO_2 is absorbed and one equivalent of O_2 developed, and for the anaerobic process in which CO_2 and two equivalents of H_2 are both absorbed.

The efficiency for O_2 production is found to be very nearly one O_2 molecule per twelve quanta absorbed. This value is obtained consistently under a rather wide range of conditions and is in agreement with values recently published from other laboratories. The efficiency for the anaerobic assimilation varies with many factors; apparently under the most favorable conditions one molecule of H_2 is taken up for approximately six quanta absorbed. Since the photosynthetic quotient CO_2/O_2 is -1 and the quotient CO_2/H_2 is $+\frac{1}{2}$, the maximum efficiency, expressed in terms of CO_2 assimilated, is the same for both the aerobic and the anaerobic assimilation.

The effects of iodine ingestion on the metabolism of normal animals. GORDON C. RING. *Department of Physiology, The Ohio State University, Columbus.*

In the hyperthyroid patient, the ingestion of small amounts of iodine usually causes the basal metabolism to drop. On the other hand, the metabolic effect of iodine in normal animals and man is very slight. In rats, supplying as much as 25 mgm. of NaI daily in drinking water over a period of three weeks, failed to stop the normal growth and did not change the basal metabolism. However, when such animals were placed in a refrigerator at 4°C., they survived for only a few days because they were unable to bring about increased thyroid activity which would elevate their metabolism. At normal temperatures, rats were able to tolerate eight times the above daily dose of NaI. This did not produce a lowering of metabolism unless there was a loss in weight. The withdrawal of iodine resulted in a subnormal metabolism which continued for three weeks or more.

Potassium chloride and pontocaine applied superficially and injected deeply into ventricular muscle bands. JANE SANDS ROBB, M. S. DOOLEY (by invitation) and ROBERT C. ROBB (by invitation). *Department of Pharmacology, College of Medicine, Syracuse University, Syracuse, N. Y.*

The two superficial muscle bundles which encompass the entire heart are readily available for surface studies and in their deep papillary portions can be impregnated without involving more than one muscle at a time. Near the Pulmonary conus there are fenestrae in the superficial sinospiral

through which the deep sinospiral can receive surface application. Electrocardiograms were taken in dogs during such surface application and after recovery, also during and after deep injections. The injected fluid was colored with India ink so that at autopsy the area injected could be identified. Three leads were recorded simultaneously. Control records where normal saline was injected produced minimal shifts of the S-T segment which rapidly disappeared. When either M/5 KCl or 0.5 per cent Pontocaine were applied to the surface by bits of filter paper, or injected deeply into a muscle bundle, the same S-T shifts characteristic of anemic infarcts in the same muscles were found. Whenever, at autopsy, the pigment was localized to one muscle alone, the magnitude of the S-T shift was greater for the deep injection than for the surface application. When more than one muscle was infiltrated, the S-T shift was sometimes less in amplitude than that obtained by surface application to a single muscle band. The deep bulbospiral muscle never approaches the surface. Injections into this muscle produced maximal upward shifts of the S-T segment in all three standard leads. In work now in press elsewhere it is reported that undue stretching of these muscles by increasing the aortic pressure also produces the S-T shifts previously reported as characteristic of lesions in these muscle bands. Hence by three types of experiment, muscle bundle localization has been demonstrated. It is thought that injection into a deeper portion of a muscle is more effective since there (e.g., in the papillary muscles) the conducting pathways are massed into a smaller cross-sectional area. If sufficiently large surface areas are treated, and the same number of pathways intercepted here also very large amplitudes of S-T displacement can be obtained.

Some reactions of curare in liquid ammonia. RICHARD G. ROBERTS, A. WM. JACKMAN and ROY A. HECHT (introduced by L. B. Nice). *Department of Physiological Chemistry, Chicago Medical School, Chicago, Ill.* (Read by title.)

In previous work (Am. J. Physiol. **119:** 391, 1937) it was shown that liquid ammonia can be used for the fractionation and conjugation of biological substances such as adrenaline. This work has been extended to the study of curare.

Curare (Merck) was not inactivated by contact with liquid ammonia for 48 hours, and after four 100 cc. extractions on a 0.5 gram sample one-half of it had passed through a 3-G sintered glass filter. All filtrations were carried out in a closed system of Dewar flasks to exclude air and moisture. When 1 gram of glycine was added to 0.5 gram of curare in liquid ammonia a vigorous exothermic reaction was started that lasted for 3 hours, and practically all of the material passed the 3-G filter on one extraction. The fraction of curare soluble in liquid ammonia forms a black and shiny plate on the wall of the Dewar flask while the insoluble fraction remains as an amorphous, very porous, tan-colored powder.

In water curare forms a coarse suspension, but curare treated first with liquid ammonia forms an extremely fine dispersion in water and after treatment with both liquid ammonia and glycine it forms an aqueous solution. All fractions were found to be biologically active although the activity varied greatly among the different fractions. The bio-assay was made upon frogs.

An anomaly in the hemoglobin absorption spectrum produced by suspensions of red cells. ELLIS J. ROBINSON (introduced by Eric Ponder). *American Cyanamid Co., Stamford, Conn.*

Using a Hardy-General Electric recording spectrophotometer, the absorption spectrum of hemoglobin, both in solution and in suspensions of red cells, has been investigated over a wavelength range of from 4000 to 7000 A. When a solution and a suspension contain the same total amount of hemoglobin, the latter, as a result of light lost through scattering, absorbs more strongly than the former between 4600 and 7000 Å. Over this wavelength range the light lost through scattering by the suspension is constant, and when due correction is made for this loss, the molar absorption coefficients of the hemoglobin in solution and the hemoglobin in the cells are the same. Below this wavelength, even when allowance is made for scattering losses, the absorption coefficient of the hemoglobin within the cells decreases sharply, until, at 4140 Å (the Soret Band) its value is only one-half of that of the hemoglobin in solution. Below 4140 Å it commences to rise again and at 4000 Å, the lowest wavelength which has been investigated, it is still rising (perhaps approaching unit again as a final value).

Variations in the pH of the suspending medium between 6.0 and 7.8 have no effect on the magnitude of these changes. The changed absorption coefficient is still observed when the hemoglobin is changed to methemoglobin by the addition of sodium nitrite or to carboxyhemoglobin by passing carbon monoxide through the initial blood, although in these cases the position of the absorption maximum is slightly shifted.

The observation of Adams, Bradley and Macallum (Biochem. J. **28**: 482, 1934) that the Soret band is absent from the absorption spectra of suspensions of red cells has not been confirmed.

Metabolic adaptations to exhausting work as affected by training. S. ROBINSON (introduced by Paul M. Harmon). *Department of Physiology, Indiana University, Bloomington.*

Nine previously untrained college men were trained for middle distance running during a period of 28 weeks. Timed races on the track were held each week and showed consistent improvement in the running ability of the men. Exhausting runs of 3 to 5 minutes duration on a motor driven treadmill were repeated at intervals of 2 to 3 weeks during the training period. In the treadmill experiments the average maximal O_2 consumption increased gradually from 52.8 to 60.2 cc. per kgm. per minute during training. Sugar in venous blood drawn 5 minutes after the exhausting runs on the treadmill was markedly elevated above basal values but showed no change attributable to training. Blood lactic acid after the runs increased gradually from an average of 13.2 mEq. per liter before training to 18 at the end of training. During work the apparent elevation of R.Q. above unity was associated with the rate of lactate accumulation. Its negative correlation with the time required to exhaust the men was therefore greater than its direct correlation with the final lactate values attained. The increased ability of the men to accumulate lactate in work was accompanied by corresponding decreases in alveolar CO_2 tension during work and in alkaline reserve of the blood after work. Basal alveolar CO_2 tension and alkaline reserve were unaffected by training. In corresponding basal and work

bloods the increases in lactate were uniformly greater than the decreases in alkaline reserve.

The effect of indole-3 acetic acid on tumor respiration. T. W. Robinson and A. B. Taylor (introduced by F. R. Steggerda). *Departments of Zoology and of Physiology, University of Illinois, Urbana.*

The respiration of fine suspensions of both spontaneous and transplanted mouse tumors in 0.9 per cent saline and in saline to which various concentrations of indole-3 acetic acid had been added was measured with the Barcroft-Warburg respirometers.

Relatively large variations in the rate of oxygen consumption of the various tumor suspensions occurred and these could be correlated in part with the H-ion concentration of the suspension. The oxygen uptake per gram per hour of the transplanted sarcoma was considerably less than that of the spontaneous mammary carcinomas. The oxygen uptake of the sarcoma increased when the suspension was more alkaline.

Inhibition of normal respiration of both types of tumor always occurred after the addition of indole-3 acetic acid. The degree of depression depends upon the concentration of indole-3 acetic acid added and even more upon the actual concentration of the active acid present at any given pH. The indole-3 acetic acid is apparently effective only in the undissociated acid form. The inhibition is a hyperbolic function of the concentration of the active acid. We used a maximum concentration of 7.45×10^{-3} mg./ml. (4.52×10^{-8} moles) of active indole-3 acetic acid and obtained a maximum inhibition of 42 per cent of the respiration in the controls.

The blood pressure response to renin and angiotonin in normal and nephrectomized dogs.[1] S. Rodbard[2] (introduced by L. N. Katz). *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.*

Recent studies have indicated that the chemical mediator of renal hypertension is destroyed at a rapid rate only by the metabolic processes occurring in the kidney; extrarenal tissues have comparatively little effect in eliminating the effect of the mediator. Since hypertension is dissipated more rapidly after removal of the ischemic kidney when a normal kidney remains than in animals with no remaining kidney tissue, a method can be established for the evaluation of various agents suggested as possible factors in the production of renal hypertension. In other words, the mediator of renal hypertension must have a more prolonged action in totally nephrectomized dogs than in those with remaining normal kidney tissue.

Blood pressures were determined on 20 trained unanesthetized dogs with the Hamilton manometer. Nephrectomies were performed using aseptic technique. Renin prepared according to the techniques of Landes and of Helmer and Page, and angiotonin prepared according to the method of Page and Helmer were used.[3] The first injection was made several hours before total nephrectomy, injections of a similar amount of the agent were made at 24 hour intervals, and blood pressures were photographically recorded until the pressor effect was dissipated. Although both systolic

[1] Aided by the A. D. Nast Fund for Cardiac Research.

[2] Eli Lilly Fellow.

[3] We are grateful to Dr. I. H. Page for supplies of renin and angiotonin.

and diastolic pressures were recorded, particular emphasis was placed upon the latter since it is a better indicator of peripheral resistance.

Injections of renin into seven dogs resulted in a similar response in terms of intensity and duration of pressor action before and after nephrectomy. In three other dogs after nephrectomy the intensity of response as measured by the height of the blood pressure level was not increased, while the duration of the pressor action was prolonged.

Injection of angiotonin into eight dogs resulted in a similar response before and after nephrectomy. In two other dogs an increased intensity and duration of the pressor effect after nephrectomy was observed.

These results fail to support the view that renin and angiotonin are agents directly responsible for renal hypertension.

Some properties of striated muscle revealed by veratrine. A. Rosenblueth, H. Hoagland and J. H. Wills (by invitation). *Harvard Medical School, Boston, Mass.*

The slow potentials of striated muscle may be associated with propagation of muscle impulses or with contraction. The relations between muscular conduction and contraction are an open question.

With progressively larger doses of veratrine the spike potential and the residual negativity increase; the mechanical response first increases, then decreases considerably. The increased contraction is due to repetitive bursts of spikes.

With small doses the first spike is normal, the residual negativity moderate, and the tetanic contraction markedly increased. With large doses the first spike is increased, the residual negativity may attain over 100 per cent of the spike amplitude, and the tetanic tension, although prolonged, may be less than that of the original twitches.

The discrepancy between residual negativity and tension indicates that the negativity is largely a conduction phenomenon—i.e., a negative after-potential. The discrepancy between spike and tension supports the conclusion that conduction and contraction are independent.

The effect of thymoxyethyldiethylamine on various pain thresholds with special reference to referred pain. Sol Roy Rosenthal, David Minard and Edward Lambert (introduced by G. E. Wakerlin). *Department of Pathology, Bacteriology and Public Health and Department of Physiology, University of Illinois, College of Medicine, Chicago.*

Thymoxethyldiethylamine raises the cutaneous threshold for pain in dogs to pinching, pricking and electrical stimuli. There is no loss of consciousness, slight or no ataxia, and no loss of knee, pupillary or abdominal reflexes. The threshold of the saphenous nerve trunks, as determined by the Harvard Inductorium, is not altered, while that of the mesenteric nerves is raised or abolished. In 27 dogs it was found that the electrical threshold for pain of the parietal and visceral peritoneum and mucosa of the intestine was raised or abolished following the administration of the drug and more or less paralleled the cutaneous electrical threshold for pain. These preliminary results suggest the possibility that in the dog, under the conditions of these experiments, pain produced by electrically stimulating the mesenteric nerves is referred through the skin.

Oxygen and carbon dioxide secretion in the swimbladder of the rockbass, Ambloplites rupestris, and its relation to hydrostatic pressure. HOWARD H. ROSTORFER (introduced by Gordon C. Ring). *Zoology and Entomology Department, The Ohio State University, Columbus.*

The secretion of oxygen into the swimbladder of many fishes is well established, and the secretion of nitrogen has been suggested. In order to better understand what causes gases to pass into the swimbladder, the relation between the hydrostatic pressure of the environment and the pressures of the gases in the swimbladder has been studied. The results show that oxygen and carbon dioxide pressures increase with corresponding changes of hydrostatic pressure, but that the nitrogen pressure in the swimbladder is not appreciably changed. Its presence can be accounted for on the basis of diffusion. Carbon dioxide, probably produced by the gas gland, drives oxygen out of combination with hemoglobin and thus raises its tension sufficiently to allow the oxygen to diffuse into the swimbladder. When the partial pressures of the O_2 and CO_2 in the swimbladder are plotted against the total pressure (hydrostatic plus atmospheric), each forms a curve which tends to level off at higher pressures. These curves have a definite relationship and indicate the blood origin of the two gases. During the first two hours of adjustment CO_2 and O_2 are almost equally important in bringing about an adjustment to increased hydrostatic pressure, but later, after the fish have apparently reached a state of equilibrium, CO_2 is removed while O_2 continues to increase. It is probable that the high CO_2 partial pressure, through local action, causes a dilation of the "oval" allowing CO_2 to leave, favored by a high coefficient of diffusion. At a total pressure of 2225 mm. Hg the final adjustment of the swimbladder resulted in a gaseous composition of from 1 to 2 per cent CO_2, 60 per cent O_2, and 38 to 39 per cent N_2. When the total pressure was reduced to 540 mm. Hg all gases were partially removed from the swimbladder, the final composition being 0.77 per cent CO_2, 4.2 per cent O_2 and 95 per cent N_2. The results show that oxygen is responsible for the greater portion of the adjustment to changes of hydrostatic pressure.

Relationship between calorimetric determinations of the upper extremities and the basal metabolic rates of normal subjects. GRACE M. ROTH, E. V. ALLEN (by invitation) and CHARLES SHEARD. *Department of Clinical Investigation and Division of Internal Medicine, the Mayo Clinic, and Division of Biophysical Research, the Mayo Foundation, Rochester, Minn.*

The rate of elimination of heat from the upper extremities of seventeen normal subjects was measured in a calorimeter. The group consisted of fourteen men and three women. The studies were made in a room in which the temperature was controlled at 25°C. and the humidity at 40 per cent; the water in the calorimeter was at initial temperatures of 21.5°C. or 31.5°C. respectively.

By immersion of one extremity in water at 45°C. during the period of measurement of the heat eliminated from the other extremity, vasomotor control of the blood flow of the extremities was removed and practically complete vasodilatation was accomplished.

On the same day, preceding the calorimetric studies, basal metabolic

rates were determined and measurements of the skin temperatures of the extremities were made.

A fairly close correlation was found in the various data obtained, particularly between the basal metabolic rates and the rate of elimination of heat from the upper extremity.

A comparison between the human vaginal smear assay and the urinary extract assay of estrogen. BORIS B. RUBENSTEIN and D. R. L. DUNCAN (by invitation). *Department of Metabolism and Endocrinology, Michael Reese Hospital and the Department of Physiological Chemistry, University of Chicago, Chicago, Ill.*

Vaginal smears and 24-hour urines were collected daily for a menstrual cycle of each of five patients. The urines were extracted and assayed on immature female mice by the method of Evans, Varney and Koch, for estrogen. The daily vaginal smears were prepared and studied for the cell types described previously. A scheme of estrogen equivalents was proposed: Cell type 8 equals Estrogen = 0; cell type 1 = Estrogen I; cell type 2 = Estrogen II; cell type 3 = Estrogen III; cell type 4 = Estrogen IV; cell type 5 = Estrogen IV; cell type 6 = Estrogen II; cell type 7 = Estrogen I. Smears consisting of combinations of various cell types were given intermediate evaluations. Curves are presented comparing the mouse uterine weights and vaginal smear assay which indicate the comparability of the two methods. These results further suggest that urinary excretion of estrogen is proportional to circulating estrogen. It seems that with suitable controls human vaginal smears may provide as accurate a gauge of estrogen production by a patient as assays of her urinary estrogen.

Slow potential changes in the electroencephalogram and functional states of the central nervous system. MORTON A. RUBIN. *Memorial Foundation for Neuro-Endocrine Research, Worcester State Hospital, Worcester, Mass.*

A comparison of the influence of voluntary hyperventilation and of sodium cyanide on the electroencephalograms (EEG's) of psychotic and non-psychotic individuals emphasizes the importance of the functional state of the brain in determining its response to a given variable. Hyperventilation may slow the 10-per-second rhythm in normal subjects, but it enhances that rhythm in the EEG of psychotic patients. With sodium cyanide there is an increase in the amount of 10-per-second rhythm in the patients' EEG. In stuporous and in narcoleptic patients, however, cyanide evokes regular, slow potential changes. The EEG of normal individuals is only slightly influenced by sodium cyanide, and then usually a decrease in normal activity is noted.

Further examples are found in the effects of sodium amytal and of metrazol. Massive doses of amytal which bring stuporous patients out of stupor and increase the amount of 10-per-second activity will put a normal individual, or a non-stuporous patient, to sleep and results in the appearance of slow potential changes. Psychotic patients who have recovered from their psychosis after metrazol shock therapy may show spontaneous bursts of regular, slow waves for some weeks after therapy has been discontinued, whereas no slow activity was present in the EEG in the psychotic state before therapy was started.

The phospholipid partition in fatty and cirrhotic livers.[1] SAUL H. RUBIN (introduced by Elaine P. Ralli). *Laboratories of the Department of Medicine, New York University College of Medicine, New York City.*

A comparative study of recent methods for the estimation of lecithin, cephalin and sphingomyelin has led to the adoption of an analytical scheme similar to that described by Thannhauser et al. (J. Biol. Chem. **129:** 709, 1939). Discussion of the results obtained is limited to lecithin and cephalin since sphingomyelin was found to constitute a small and constant fraction (up to 5 per cent) of the liver phospholipids in the cases detailed below. The values are expressed as percentages of the total phospholipids.

The livers of 67 normal albino rats (150–200 grams), analyzed in groups of approximately 10, contained 59 per cent lecithin (55–62) and 38 per cent cephalin (35–43). Ninety-four rats with fatty livers produced by a high-fat, low-protein diet, showed 64 per cent lecithin (58–68) and 33 per cent cephalin (28–40). These differences cannot be considered significant.

However, fatty livers obtained from 5 depancreatized dogs contained 69 per cent lecithin (57–79) and 27 per cent cephalin (17–39), as compared with 51 per cent (46–58) and 45 per cent (38–50), respectively, in 6 normal dogs. The depancreatized dogs were maintained for periods of 9 to 16 weeks, while the rats were sacrificed after 3 weeks. The low blood plasma phospholipids found in these depancreatized dogs (Am. J. Physiol. **129:** 578, 1940) were accompanied by a relative diminution in the lecithin-cephalin ratio.

Nine normal human livers contained 56 per cent lecithin (49–63) and 39 per cent cephalin (32–48). Six cirrhotic human livers with varying degrees of fatty infiltration showed the following percentages for lecithin: 62, 66, 67, 69, 74 and 92; the last was found in a liver which contained 24.4 per cent fatty acids. The cephalin figures were correspondingly reduced.

A tentative conclusion which may be drawn from these data is that in long-standing fatty infiltration of the liver and in cirrhosis, the lecithin-cephalin ratio increases, although the absolute concentrations of both are diminished.

Taste disturbances from thalamic lesions in monkeys.[2] T. C. RUCH, MARVIN BLUM (by invitation) and J. BROBECK (by invitation). *Laboratory of Physiology, Yale University School of Medicine, New Haven, Conn.*

Börnstein's "opercular theory" of the cortical representation of taste, together with the data on the origin of the thalamo-cortical projection to this region (Walker; Le Gros Clark), suggest that the taste pathways may relay in the arcuate nucleus (N. ventralis posteromedialis) of the thalamus, or adjacent nuclei. In preliminary experiments on macaque monkeys, the threshold for bitter was determined by the "preference method" and lesions were made bilaterally in the region of the arcuate nucleus with the Horsley-Clarke stereotaxic apparatus. Each of three monkeys so operated have shown nearly complete loss of taste in the early postoperative period.

[1] This research was aided by a grant from The Milbank Memorial Foundation.
[2] Aided by grants from the Fluid Research and G. H. Knight Memorial Funds, Yale University School of Medicine.

followed in the case of the first two monkeys by a recovery that was almost complete. Both monkeys exhibited some type of disturbance of the chewing mechanism and a striking hyperphagia and adiposity. In the third monkey the lesion was made slightly more rostral and superior. A severe and permanent taste deficit resulted but there was no disturbance of eating behavior. A lesion several millimeters superior to those of the first two monkeys was entirely without effect on taste ability.

Carbohydrate metabolism in the eviscerated rat.[1] JANE A. RUSSELL.[2] *Laboratory of Physiological Chemistry, Yale University School of Medicine, New Haven, Conn.*

The rôles of the anterior pituitary and the adrenal cortex in the metabolism of carbohydrate have been studied in the eviscerated rat. In the first series of experiments, the length of time of survival after evisceration, without glucose administration, and the changes in carbohydrate levels of muscle and blood during this time were compared in hypophysectomized, adrenalectomized, and control series of rats, and in hypophysectomized animals treated with saline A.P.E. or with adrenal cortical extract. In order to determine accurately changes in muscle glycogen content after evisceration, it was found necessary to remove the adrenal medullae from all animals prior to the experiments; this operation prevented the glycogenolysis in muscle which otherwise occurred as a result of the liberation of adrenalin during the experiments.

The survival time after evisceration was reduced by about a half by hypophysectomy and to a lesser extent by adrenalectomy. The muscle glycogen levels in adrenalectomized and control rats did not change during 1 hours after evisceration, but they fell considerably in the hypophysectomized animals. A.P.E. prolonged the survival of the hypophysectomized rats to the normal limits and prevented the fall in muscle glycogen. Cortin also prolonged the survival time, but it only partially, if at all, prevented the loss of muscle glycogen.

The results in hypophysectomized rats were extended by determination of the rate at which it was necessary to infuse glucose to maintain the blood sugar at a constant level after evisceration. The rate in control rats was close to 13.5 mgm. per 100 grams per hour. In the hypophysectomized rats, this rate of infusion was not nearly sufficient; the required amount varied somewhat but was about 25 mgm. per 100 grams per hour in most cases. The average rate of fall of the blood sugar level after the cessation of the infusion was 3 times as fast in the hypophysectomized rats as in the controls. The changes in muscle glycogen during infusion, and the effects of A.P.E. on the infusion rate required to maintain blood and muscle carbohydrate levels are now being studied.

Changes in palmar skin resistance associated with muscular work. A. H. RYAN and E. L. RANSEEN (by invitation). *Private laboratory, 820 N. Michigan Ave., Chicago, Ill.*

Work was done on a bicycle arranged to drive an electric generator. Load was varied by means of electric bulbs. A voltmeter indicated speed

[1] Aided by grants to Dr. C. N. H. Long from the Committee on Research in Endocrinology, National Research Council.

[2] Lalor Foundation Fellow.

of pedaling and a watthour meter recorded the amount of work done. Two shallow pans were supported in a position corresponding to that of the handle bars. In each pan was placed a large zinc plate connected in a circuit with an ohmmeter. Into the pans was placed a solution of an electrolyte. The hands were placed palms down on the zinc plates, the entire palmar surface being submerged in the solution. Observations were made under two conditions of performing work; one in which the work was continuous for a period of approximately ten minutes, and one in which there were seven work periods of one minute each with thirty seconds of rest between each work period.

Within the first minute after the subject has worked with a sufficient load at a given initial rate of speed, there is a large drop in the resistance. Then the rate of fall of resistance sharply declines and there may be an actual increase in resistance. This rise or period of relatively smaller decline is then followed by a marked increase in the rate of fall in resistance. The fall of resistance then continues, generally reaching its lowest level at the end of the work period.

When work periods alternate with rest periods, a marked increase in resistance occurs during the early rest pauses. In successive rest pauses, this rise decreases in magnitude and may be replaced by a fall.

In continuous periods of work in which the speed remains constant, the resistance is found to vary inversely with increases and also with decreases of load.

Some effects of denervation of the salivary glands. ROSALTHA H. SANDERS (introduced by A. J. Carlson). *Department of Physiology, University of Chicago, Chicago, Ill.*

Saliva was collected from a common fistula of Wharton's and Bartholin's ducts on the denervated side.

Two dogs after chorda-lingual section on one side showed the following changes:

1. A slow secretion, sometimes discontinuous. A larger reflex secretion accompanying the ingestion of food. In one dog, this reflex secretion amounted to about one per cent of the preoperation rate after four days, then after some five months equalled eighteen per cent of the preoperation reflex rate. In the other dog, the reflex saliva has amounted to about twelve per cent of the preoperation rate from the sixth to the hundredth day of observation.

2. A rise in permeability to glucose with the appearance of sugar in the saliva at a blood sugar level of 200 milligrams per cent. With a normal gland, sugar appears in saliva only at a blood sugar level of 300 milligrams per cent.

3. No significant change in composition of reflex saliva of the paralytic glands in regard to mucin, total solids and total chlorides, except a fall in chlorides after chorda section followed by a rapid return to the preoperative level.

One dog has been observed for nine months after sympathectomy of the submaxillary and sublingual glands on one side, as follows:

1. A progressive decrease in rate of reflex secretion to two thirds of the normal rate at the end of three months, followed by a rise to almost the normal rate after nine months. No continuous secretion.

2. No other changes.

One dog with a chorda section followed in four days by an ipsilateral sympathectomy of the salivary glands, is now under observation, with the following results:

1. No reflex secretion. A slow secretion which is not continuous (sometimes, the opening of the fistula is dry).

2. Saliva collected after pilocarpine injection contains appreciable amounts of glucose at a blood sugar level of 200 milligrams per cent, indicating an increased permeability to glucose in the completely denervated glands similar in degree to that seen in the parasympathectomized glands.

Gastric secretion during the night in normal individuals and peptic ulcer patients. D. J. SANDWEISS, M. H. SUGARMAN and M. H. F. FRIEDMAN (introduced by T. L. Patterson). *Harper Hospital and Wayne University College of Medicine, Detroit, Mich.*

Normal individuals and patients with duodenal ulcers were given evening meal (at 6 p.m.) consisting of beef extract, white fish, tea, crackers, tomato juice and buttered toast. Hourly aspirations were made from 6 p.m. to 8 a.m. The stomach was completely emptied at midnight and again hourly until 8 in the morning.

1. In both normal individuals and ulcer patients, two peaks in concentration of free acid usually occurred; one peak, between eleven p.m. and one a.m., and the other between three a.m. and five a.m. After five in the morning both the volume of secretion and the concentration of free acid usually were very greatly reduced, the juice often showing no free acid.

2. The volume of secretion was significantly higher in the ulcer patients as compared with the normal individuals, though the concentrations of free acid in the two groups were approximately the same.

3. Bile was frequently present, usually after midnight. Mucus was often found, particularly towards the morning hours.

4. In both our normal individuals and ulcer patients, the volumes secreted were much higher than others have reported. However, this increase in volume may be explained by the fact that we used beef extract and fish in our meals,—foods known to be strong stimulants of gastric secretion.

Human skin reactions resulting from intracutaneous injection of animal blood plasmas and their alteration by bacterial action. GEORGE M. SAVAGE (by invitation), HENRY LONGSTREET TAYLOR (by invitation) and ANCEL KEYS. *Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis.*

Intracutaneous skin tests (5,160) were applied to human subjects (440). Materials used were sterile blood plasmas, diluted 1 to 70, from cow, goat, sheep, horse, hog, fox, rabbit, guinea pig and human. The tests were read at 20 minutes, 2 hours and 24 hours and were graded on a scale of 1 to 4 compared with isotonic phosphate buffer control. When sterile plasmas were used the following positive reactions were observed at 20 minutes: bovine plasma, 10 per cent of 250 individuals; goat plasma, 10.7 per cent of 250 individuals; rabbit plasma, 16 per cent of 50 individuals; hog plasma, 41 per cent of 100 individuals; dog plasma, 34 per cent of 50 individuals; horse plasma, 20 per cent of 100 individuals; sheep plasma, 21 per cent of 150 indviduals; guinea pig plasma, 4 per cent of 50 indi-

viduals; fox plasma, 40 per cent of 50 individuals, and human plasma, 12 per cent of fifty individuals. Occasional individuals were sensitive to most of the several plasmas but, in general, cross sensitivities were infrequent. For example, of 100 persons 10 were sensitive to bovine plasma and 6 to goat plasma but only 2 of these persons were sensitive to both.

In general, sterile plasmas which previously had been slightly contaminated and stored in the cold elicted additional reactions of 2 types: 1, reactions at 20 minutes which became negative in 2 hours; 2, a delayed reaction appearing at 2 hours and persisting for 24 hours. Such beef plasmas gave, on the average, 8 times as many positive reactions at 20 minutes as plasma of completely sterile history but attempts to isolate the responsible organisms failed. Some of the organisms producing the delayed reaction have been isolated. When these were cultured for 8 hours at 25°C. on relatively non-reactive beef plasma this plasma, after sterilization by Seitz filtration, produced the delayed reactions in almost 100 per cent of test cases.

Experimental ulceration of the gastro-intestinal tract. M. J. SCHIFFRIN (introduced by B. P. Babkin) and ALTHEA A. WARREN (by invitation). *Department of Physiology, McGill University, Montreal, Canada.*

A method for the production of jejunal ulcer in acute experiments by perfusion with a solution of pepsin in HCl was previously described (Schiffrin, Proc. Soc. Exper. Biol. Med. **45:** 592, 1940). Further studies utilizing this method have been made. Segments of small intestine were perfused with *a*, HCl, pH 1.0–1.2; *b*, HCl and pepsin, pH 1.0–1.2; *c*, HCl and pepsin in various proportions; *d*, HCl, pepsin and either colloidal aluminum hydroxide (pH 3.5) or aluminum phosphate gel (pH 2.0). Perfusion was carried out at body temperature at a rate of 1–2 cc. per minute. Each segment of intestine was provided with a mesenteric pedicle of adequate size. Acid *per se* produced necrosis and damaged the villi. Acid and pepsin ulcerated and in some cases perforated the jejunum within 12 hours. The addition of either of the aluminum compounds to the acid-pepsin solution prevented any damage to the small intestine.

The same method has been applied to the stomach and duodenum. In these experiments the pancreatic and biliary ducts were ligated. Acid damaged the stomach and duodenum much more than the jejunum. This may be attributed in part to the presence of pepsin in the stomach, since it has been shown that acid on leaving the stomach possesses peptic activity. Perfusion with acid and pepsin resulted in damage to the stomach in the region of the lesser curvature and profuse ulceration of the duodenum.

Apparatus for determining the tensile strength of rat tibiae. A. A. SCHILLER, (by invitation), H. C. STRUCK and C. I. REED. *Department of Physiology, University of Illinois, College of Medicine, Chicago.* (Demonstration.)

This apparatus is a small scale modification of equipment used by engineers for testing the tensile strength of structural materials. Graphic recording of deflection stress and breaking weight makes possible quantitative comparison bones from rats subjected to various dietary modifications.

A study of the comparative tensile strength of the tibiae of normal and healed rachitic rats. A. A. SCHILLER (by invitation), H. C. STRUCK and C. I. REED. *Department of Physiology, University of Illinois, Chicago.*

The elastic properties of bone are disturbed in rickets as evidenced by breaking load and deflection stress. This disturbance is directly related to the number of days on the rachitogenic diet and to the time on the curative diet. The most significant difference between control and experimental animals is found up to 42 days, after which the healing process is sufficiently advanced to indicate that the tensile strength of the bone is not permanently decreased by a rachitogenic regimen of 41 days duration.

Anoxemic hyperpnea in the dog. C. F. SCHMIDT, P. R. DUMKE (by invitation) and H. P. CHIODI (by invitation). *Laboratory of Pharmacology, University of Pennsylvania, Philadelphia.*

In 14 vagotomized dogs, lightly anesthetized with morphine and chloralose, pulmonary ventilation was measured during the following sequence, arterial blood being collected for estimation of pO_2, pCO_2, and pH during the steady states so produced: inhalation of room air, of 100 per cent O_2, of 3.5 per cent CO_2 in O_2, of room air, of 10–12 per cent O_2 in N_2, and of 10–12 per cent O_2 plus 3.5 per cent CO_2; the sequence was repeated after complete carotid denervation save for the use of 12–14 per cent O_2, which now produced as severe anoxemia as 10–12 per cent had done previously. It was found that: 1. inhalation of pure O_2 often caused a decrease in pulmonary ventilation before denervation but within 2 minutes the previous level was always regained without any change in arterial pCO_2; after the denervation inhalation of O_2 consistenly stimulated breathing. 2. Simple anoxemia regularly caused hyperpnea before denervation but afterward it was only depressant; the threshold for anoxemic hyperpnea lay at an arterial pO_2 of about 55 mm. Hg. 3. Addition of CO_2 to the anoxic gas mixture before denervation elicited a greater hyperpnea than either anoxemia or hypercapnia alone, but after denervation the response to CO_2 in O_2 was consistently greater than that to CO_2 with low O_2. 4. The drop in pO_2 between the inspired air and the arterial blood (ΔpO_2) was diminished by the addition of CO_2 to an inhaled mixture low in O_2, as by increased depth of breathing however produced; chemoreceptor reflexes were responsible for by far the largest part of this phenomenon, changes in the dissociation curve of oxyhemoglobin being relatively unimportant. These results indicate that 1, no important amount of chemoreceptor activity was maintained in these animals by the arterial pO_2 associated with quiet breathing of room air; 2, the hyperpnea of anoxemia was entirely due to chemoreceptor reflexes, the only effect of anoxia directly on the center being to depress its response to CO_2; 3, the hyperpnea of mild hypercapnia was not demonstrably dependent on reflexes.

Effect of insulin and thyroidectomy on glucose tolerance in toxic goiter. C. R. SCHMIDT, W. S. WALSH (by invitation) and V. E. CHESKY (by invitation). *Department of Surgery, Hertzler Clinic, Halstead, Kansas.*

Diminished glucose tolerance in thyrotoxicosis has been attributed to a variety of causes including 1, inability of the liver to store glucose; 2, increased hepatic glycogenolysis; 3, insulin deficiency or refractivity,

and 4, increased rate of absorption from the intestine. Certain of our observations suggest that increased gluconeogenesis may be a contributing factor.

Glucose tolerance and utilization was studied before and after thyroidectomy in thirty patients with toxic goiter and in four patients with non-toxic goiter. In the subjects with toxic goiter, glucose (1.75 grams per kilo of body weight) administered orally or intravenously at a rate of 0.75 gram per kilo per hour gave characteristic blood sugar curves (capillary blood method) with abnormally high peaks maintained over prolonged periods. Glucosuria occurred in all instances. From five to fifteen grams of glucose were recovered in the urine. Insulin, one unit per five grams of glucose, added to the intravenous glucose solution eliminated the glucosuria.

When the same quantity of glucose was administered intravenously to thyrotoxic subjects during a twenty minute period an entirely different type of blood sugar curve was obtained. Under these conditions blood sugar concentration reached a peak at thirty minutes and then declined rapidly to subnormal levels (3 hrs.). Insulin produced a quantitative but no qualitative change in the blood sugar curves. The quantities of glucose excreted in the urine were essentially the same as when the glucose was administered intravenously, with and without insulin, at a rate of 0.75 gram per kilo per hour. It is felt that increased gluconeogenesis is an important factor in the altered carbohydrate metabolism associated with thyrotoxicosis, and that the blood sugar response to excessive amounts of glucose is influenced appreciably by diminished gluconeogenesis.

Glucose tolerance was restored to essentially normal within five days after bilateral thyroidectomy in toxic goiter patients. Hemi-thyroidectomy ameliorated but did not correct the disturbance in glucose tolerance. Thyroidectomy produced no alteration in the normal glucose tolerance of patients with non-toxic, nodular or non-toxic colloid goiters.

Oxygen therapy in shock. J. G. SCHNEDORF and T. G. ORR (introduced by A. C. Ivy). *Department of Surgery, University of Kansas School of Medicine, Kansas City.*

This work was done to determine the effect of continuous inhalation of high oxygen concentrations upon shock due to trauma, histamine and hemorrhage.

While the degree of trauma, as indicated by the average amount of fluid loss into the injured limb, was the same in 10 control dogs breathing atmospheric air (4.26 per cent total body weight) and the 10 dogs treated with 100 per cent oxygen inhalations (4.11 per cent total body weight) the average length of life of the control dogs was 4.5 hours and those treated with oxygen was 7.7 hours. Thus, oxygen therapy caused a 70 per cent increase in the length of life of dogs in traumatic shock. The blood pressure of the dogs treated with oxygen was higher 4, 6 and 8 hours after the trauma than the untreated group. Oxygen increased the length of life of 10 dogs in histamine shock (blood pressure maintained at 40 mm. of mercury) by 71.7 per cent. The control dogs lived 5.6 hours and the treated dogs 9.8 hours. Loss of blood by repeated hemorrhage equal to 2 per cent of the total body weight in the 10 control dogs caused a 54 mm. of mercury fall in blood pressure and only a 35 mm. fall in the 10 dogs

treated with oxygen. After a blood loss of 4 per cent of the body weight, the blood pressure of the control dogs was 35 mm. and of the treated dogs 39 mm. of mercury. Oxygen therapy enabled the treated dogs to tolerate a 15 per cent greater blood loss (4.82 per cent body weight) than the control dogs (4.19 per cent body weight). It also resulted in a 17 per cent increase in the length of life of the treated dogs (3.60 hrs.) over that of the control dogs (3.08 hrs.). Increased hematocrit readings, indicating hemoconcentration, were observed in all three types of shock. A decrease in the oxygen content and saturation of blood from the femoral artery and vein was observed in nembutalized dogs in traumatic shock. Oxygen therapy significantly increased the oxygen in the arterial and venous blood.

Inhalation of high oxygen concentrations has a beneficial action upon shock due to trauma, histamine and hemorrhage.

Some effects of insulin and glucose on fasting external pancreatic secretion. V. BROWN SCOTT, U. J. COLLIGNON and H. J. BUGEL (introduced by P. M. Harmon). *Department of Physiology, Indiana University School of Medicine, Bloomington.*

We were unable to observe consistent alterations in the volume and proteolytic activity of pancreatic juice collected at half-hour intervals before and after injections of insulin and glucose in fasting dogs which secreted large quantities of pancreatic juice (Am. J. Physiol. **129:** P457, 1940). Since a temporal correlation between hunger motility and pancreatic secretion has been observed (Scott and Bugel, Am. J. Physiol. **131:** 60, 1940) and since many investigators have observed gastric hypermotility with insulin, we decided to investigate the action of insulin and glucose upon gastric motility and pancreatic secretion in the fasting dog.

Observations were made on dogs with pancreatic fistulae (Inlow) and gastrostomies (Carlson) which were trained to lie on a padded table after a 24 hour fast. Pancreatic secretion was recorded by a drop recorder and gastric motility was registered by the usual intragastric balloon and water manometer technique.

Subcutaneous insulin (1 unit per kgm.) elicited, within 50 to 60 minutes, an increased flow of pancreatic juice and gastric hypermotility. Intravenous insulin produced an initial inhibition of pancreatic secretion and gastric motility which was followed by an augmentation of the secretion and motility when hypoglycemia (25 to 40 mgm. per cent) appeared. Intravenous glucose (1 gram per kgm.), administered as a 20 per cent solution during the insulin hypoglycemia, resulted in a transient inhibition of pancreatic secretion and gastric hypermotility.

In dogs having bilateral vagotomies, the gastric hypermotility of hypoglycemia was abolished but the pancreatic augmentation remained unaffected. Vagotomy did not remove the initial inhibitory action of intravenous insulin on pancreatic secretion and the inhibitory action of glucose also remained unaffected.

A colorimetric redox method for the determination of vitamin K_1 and similar quinones. JOHN V. SCUDI (introduced by Hans Molitor). *Merck Institute for Therapeutic Research and Research Laboratories of Merck & Co. Inc., Rahway, N. J.*

A colorimetric method for the determination of vitamin K_1 and asso-

ciated quinones has been devised on the basis of the redox titrations of Trenner and Bacher (J. Biol. Chem. **137:** Feb. 1941). The method involves a catalytic reduction of the quinone in butanol in the presence of phenosafranin as the indicator. The resulting hydroquinone is then treated with an excess of a butanol solution of 2,6-dichlorophenol-indophenol in the absence of air. A partial reduction of the indophenol to the leuco base results. This reduction in the color of the indophenol is a measure of the quinone originally present. It is measured in the Evelyn colorimeter, at 650 mμ within 2 to 3 minutes after mixing solutions of the hydroquinone and the 2,6-dichlorophenol-indophenol. Taking readings in this short interval of time diminishes the influence of extraneous reducing substances. The test is applicable to solutions containing 2 to 10 micrograms of 2-methylnaphthoquinone per cubic centimeter of solution.

Application of the method to highly colored petroleum ether extracts requires the removal of these colored impurities. This was effected by reducing the extract over Raney nickel in the presence of an excess of phenosafranin. The hydroquinone was extracted with Claisen's alkali, and the interfering colors were removed by washing with petroleum ether. The sodium salt was hydrolysed and the hydroquinone was extracted with petroleum ether. Colorless solutions were obtained. These were oxidized to the quinone, and the test was performed as above.

Data obtained by the application of the method to a number of foodstuffs, tissues, etc. are presented. The method is also applicable to vitamin E.

Survival of the respiratory (gasping) mechanism in young animals. W. A. SELLE and T. A. WITTEN (by invitation). *Department of Physiology, University of Texas School of Medicine, Galveston.*

Several observers have reported that young animals of certain species are less susceptible to asphyxia than adults. Studies were undertaken to determine whether the primitive respiratory mechanism (gasping) itself survives longer in the young than in the old.

Animals of various species (rats, mice, rabbits, cats and dogs), varying widely in age, were subjected to anoxic or asphyxial conditions by: 1, breathing nitrogen, CO_2, illuminating gas; 2, ligation of cerebral vessels; 3, decapitation. Gasping movements of the mandible were recorded mechanically. It was found that the survival of gasping in animals ranging from one day to slightly past weaning is inversely proportional to age. Thus, a one-day old rat gasps from forty to eighty times during a period of thirty minutes or more following complete ligation of the cerebral vessels or isolation of the head; a six-weeks old animal usually gasps five to eight times and all activity ceases within fifteen to thirty seconds. Information concerning the essential factors responsible for this difference is lacking although metabolism differences appear to be involved. The survival of the gasping mechanism parallels other physiological processes as it was further found that: 1, pupillary responses of the isolated head; 2, trunk reflexes of the spinal animal; 3, action of the exposed heart, are all retained longer the younger the animal.

The influence on the survival of the gasping mechanism of temperature, metrazol, acetylcholine, insulin, various anesthetics and decerebration will be discussed.

The anesthetic effect of steroid hormones. HANS SELYE. *Department of Anatomy, McGill University, Montreal, Canada.* (Motion picture demonstration.)

The anesthesia produced by progesterone is demonstrated in this colored film as an example of a typical steroid hormone anesthesia. In the experiments illustrated in this film, the anesthesia was induced in rats by the intraperitoneal administration of 2–5 mgm. of progesterone in peanut oil. The second part of the film shows the performance of a sub-total hepatectomy in the rat using only progesterone as an anesthetic. Desoxycorticosterone acetate and various other steroid hormones proved to exert a similar anesthetic action when administered by the intraperitoneal route. Pretreatment with atropin prevents, while vagotomy actually intensifies the anesthetic action of progesterone.

Oxygen utilization by starfish eggs. HERBERT SHAPIRO. *Marine Biological Laboratory, Woods Hole, Mass., and Vassar College, Poughkeepsie, N. Y.* (Read by title).

The oxidative activity of eggs of the starfish, *Asterias forbesii*, obtained in the early summer, was measured in Warburg manometers over a temperature range 11.5°C. to 27.5°C. Unfertilized eggs showed a constant rate of uptake for periods as long as ten hours, whereas fertilized eggs respired at a relatively constant rate at first, and then exhibited a gradually increasing rate as embryological development proceeded in the respirometers. On fertilization, the change in rate is variable in different experiments. There may be no alteration, or even a slight drop, but in most of the observations, an average increase of roughly 30 to 50 per cent appeared. This statement respecting the influence of fertilization on respiratory activity applied, on the whole, throughout the temperature range employed, with a tendency for the average ratio, fertilized rate/unfertilized rate (F/U), to decline slightly with elevation of temperature. Starfish eggs are not as uniform as sea urchin (Arbacia punctulata) eggs with respect to size, configuration, maturity, cleavage and other characteristics. This non-uniformity may well account for the observed variations in F/U.

In the sea urchin egg, the value of F/U is uniquely dependent upon the temperature (Rubenstein and Gerard, 1934) and varies inversely with temperature. This is in marked contrast to the starfish, where the temperature dependence of F/U is negligible. An F/U temperature dependence argues for a qualitatively different oxidative system or systems in the fertilized egg, which either replaces or is added to that of the unfertilized cells. The sea urchin egg is known to demonstrate a cyanide-sensitive system upon fertilization, which is in addition to the non-cyanide sensitive system characteristic of the unfertilized egg. Two alternative inferences are possible from these experiments, though no direct chemical proof is given for either: *a*, in *Asterias*, either no new oxidative system is brought into play on fertilization, or if one does appear, its contribution to the overall oxygen uptake is small; *b*, if a new system is introduced on fertilization, which makes a major contribution to the observed oxygen uptake, its temperature dependence function is similar to that of the system or systems prevailing in the unfertilized egg.

The blood precursors of the short chain fatty acids of milk. J. C. SHAW and C. B. KNODT (introduced by M. B. Visscher). *Department of Dairy Industry, Storrs Agricultural Experiment Station, Storrs, Conn.*

Arteriovenous differences demonstrated that acetone bodies were used by the lactating gland of the cow. Fractionations disclosed that the utilization of acetone bodies was limited to β-hydroxybutyric acid. In 16 experiments there was a mean utilization of 2.49 ± 0.325 mgm. per cent β-hydroxybutyric acid. According to our arteriovenous difference figures the quantity of β-hydroxybutyric acid utilized is just sufficient to provide for the fatty acids C_{14} and lower. Such synthesis would explain the high R.Q. of the lactating gland. Approximately 40 per cent of the total oxygen consumption of the gland would be required for the complete oxidation of β-hydroxybutyric acid for energy purposes. Considering the high R.Q. of the normal lactating gland such a postulation does not appear to be warranted.

The R.Q. of the lactating gland in periods of inanition and cod liver oil feeding was less than unity. Likewise marked decreases in Reichert-Meissl values of the milk fat occurred in both cases. Simultaneously the β-hydroxybutyric acid arteriovenous differences decreased significantly. When dextrose was fed in large quantities or pumped into the rumen there was a fall in blood acetone bodies of over 50 per cent which resulted in a decrease in the arteriovenous difference of β-hydroxybutyric acid. Coincident with the decline in blood β-hydroxybutyric acid there was a marked decrease in the saponification number, Reichert-Meissl value and Polenske value of the milk fat.

In severe ketosis in dairy cows in which both blood glucose and blood lactic acid were less than 50 per cent of normal, the R.Q. of the active gland was in excess of unity and indicated that carbohydrate material is probably not used for fat synthesis in the gland.

It is concluded that β-hydroxybutyric acid is probably used in the synthesis of the short chain fatty acids of milk.

Dark adaptation: surveys of normal subjects and clinical applications. CHARLES SHEARD, HUGO L. BAIR (by invitation) and LOUIS A. BRUNSTING (by invitation). *Division of Biophysical Research, The Mayo Foundation, and Sections on Ophthalmology and on Dermatology, The Mayo Clinic, Rochester, Minn.*

Data will be presented on a group of normal persons emphasizing the importance of intensity of and duration of exposure to light, the size and location of area of retinal stimulus, and the color of the stimulus in the determination of levels of cone-rod adaptation. Measurements of visual adaptation during the course of 20 to 30 minutes were made on 60 pilots of a commercial air line and on 110 school children between the ages of eight and fourteen. Twenty of the group of school children were examined on several occasions and with different standards of light adaptation and sizes of retinal stimulus area. These data have been examined statistically and the probable spread of 70 to 80 per cent of the values of normal dark adaptation determined. Methods and standards will be suggested for selection and rejection for special services. These data and conclusions drawn from them will be compared with the findings in certain pathological conditions.

The alleged antithyroid action of vitamin A. R. F. SHEETS (by invitation) and H. C. STRUCK. *Department of Physiology, College of Medicine, University of Illinois, Chicago.*

Vitamin A fed in large, but non-toxic, doses to rats has been found to cause a temporary reduction in the metabolic rate. This reduction was present in normal, thyroid-fed, as well as in thyroidectomized animals. The temporary nature of the action, and particularly the fact that the action is similar in normal and thyroidectomized rats had led to the conclusion that the thyroid gland is not concerned.

Chloride space in hypertrophied hearts of hyperthyroid rats. WALTER B. SHELLEY (by invitation) and CHARLES F. CODE. *Department of Physiology, University of Minnesota, Minneapolis.*

These experiments were designed to ascertain whether or not the relative size of the chloride space of the cardiac muscle of rats undergoes changes during the hypertrophy associated with thyroid feeding. All animals received a standard laboratory diet of fox chow. Three series of rats have been studied, a control series receiving only the standard diet and two groups receiving the standard diet plus desiccated thyroid gland. The diet of group 1 of the thyroid-fed animals contained 0.7 per cent thyroid and this was administered for 7 days. Group 2 received 0.7 per cent thyroid in the diet for 23 days and then 0.3 per cent for a further 22 days. At the end of the 7 and 45 day periods chloride determinations were made on the plasma (Van Slyke) and heart (Sunderman and Williams) of each rat. The heart chambers were opened and all external blood carefully removed from the surfaces. Calculations of chloride space were made according to Hastings and Eichelberger (*J. Biol. Chem.* **117:** 73, 1937) and results expressed as grams per 100 grams of whole fresh heart. It should be pointed out that the chloride space expressed in this manner is not on the basis of blood- and fat-free tissue. No significant difference was found between the chloride space of the normal and the hypertrophied hyperthyroid hearts (table).

	NO. OF RATS (ALL ♀)	WEIGHT RANGE	HEART WEIGHT RANGE	CHLORIDE SPACE GRAMS/100 GRAMS WHOLE FRESH HEART	
				Range	Average
		gms.	*gms.*		
Controls	15	200–255	0.53–0.67	21.1–25.9	23.2
Group 1	5	200–230	0.76–0.87	21.7–24.0	23.1
Group 2	9	170–200	0.77–0.98	21.0–26.8	24.8

The effect of glutathione on somatic growth of rats. T. R. SHERROD (by invitation), H. C. STRUCK and C. I. REED. *Department of Physiology, College of Medicine, University of Illinois, Chicago.*

Intraperitoneal injection of 25 mgm. of pure reduced glutathione daily into rats resulted in a definite retardation of body growth as measured by body weight and body length. The effect was more pronounced in the males.

Effect of nerve stimulation on blood flow in coronary arteries.[1] R. E. SHIPLEY (by invitation), A. ROTTA (by invitation), D. E. GREGG and W. H. PRITCHARD (by invitation). *Departments of Medicine and Physiology, Western Reserve University, Cleveland, O.*

Measurements of coronary flow were made in anesthetized, open chest dogs by use of: 1, a rotameter to obtain a general picture of events; 2, an orifice plate meter and registration of peripheral coronary pressure for flow details.

In the left coronary with essentially a constant heart rate and blood pressure, stimulation of the left stellate ganglion or its cardiac fibres induces mild to large augmentations of coronary inflow (up to 350 per cent with rotameter) which may persist 10 to 12 minutes. Both systolic and diastolic inflows increase (orifice meter) especially at the points on the flow curve which probably represent largely intramural flow. When the flow increases are sizeable, both systolic and diastolic pressures decrease thus indicating coronary dilatation. In those experiments in which the blood pressure is also elevated, the peripheral coronary pressure decreases and the flow increases on an average 600 per cent more than it does with a comparable elevation of blood pressure through aortic constriction. At times these flow increases are preceded by a few seconds of mild flow reduction, both during systole and diastole. Excision of the ganglion and nerve does not alter the flow.

To date, in the right coronary stellate stimulation and excision, and in both coronaries sciatic, sinus, and vagus nerve stimulation and excision have very little if any effect on coronary flow.

Prolongation of survival time in Mann-Williamson dogs by supplementing diet with amino acids. DAVID SHOCH and S. J. FOGELSON (introduced by K. K. Jones). *Department of Surgery, Northwestern University Medical School, Chicago, Ill.*

The development of ulcer has been delayed and the survival time prolonged in Mann-Williamson dogs by feeding of "special" diets. In general these diets have been characterized by high calorie and digestive enzyme content, as for example, a prepared dog food, as well as pancreas, liver and whole milk.

We have observed that loss of weight is usually followed by ulcer manifestations in these animals. For this reason a series of fifteen Mann-Williamson dogs were placed on a basal diet of milk, dextri-maltose, glucose, vitamins and minerals. This fairly adequate diet was supplemented with a protein hydrolysate so that the amino acid intake was one gram per kilo. The average survival time of this series was two hundred and fifteen days. A control series of ten dogs was also prepared who received the basal diet with an equivalent supplement of casein but no protein digest. The average survival time of this control series was just over a hundred days. Thus the difference in survival time of the amino acid series over the casein control series was about three months.

Gastric analyses on both series gave practically identical findings.

Although prevention of ulcer formation in Mann-Williamson dogs was not attained, a significant delay in its onset was obtained. This permitted

[1] Aided by a grant from the Commonwealth Fund.

the conclusion that improved nutrition with ease of assimilation of diet are significant factors in experimental ulcer.

Unilateral progression independent of proprioception. P. S. SHURRAGER (introduced by W. Horsley Gantt). *University of Pennsylvania, Philadelphia.*

In a previous report the author presented evidence of unilateral progression reflexes in the spinal dog. Further observation has disclosed that when the cord is ligated and transected above the 3rd lumbar spinal roots with all roots on the side stimulated cut except the 6th sensory and 7th motor, and the stimulus is maintained at maximal intensity, the development of unilateral progression is directly dependent upon the number of intact contralateral motor roots. All contralateral sensory roots can be eliminated without the appearance of unilateral progression. As the contralateral motor roots are progressively severed, unilateral progression develops from the original flexion-relaxation of the simple spinal reflex to flexion-relaxation-extension-relaxation pattern which, when all contralateral motor roots have been cut, may repeat itself as many as 3 times.

A decrease in stimulus intensity results in failure of the initial flexion-relaxation-extension-relaxation phase. All activity is inhibited for a period approximately equal to the time originally required for this first phase. At the end of the latent period the second and third phases of the unilateral progression sequence appear as usual.

These phenomena indicate that unilateral progression may primarily depend upon a fixed neural pattern which is not dependent upon the action of proprioceptive impulses.

The relative elasticity of the sarcolemma and of the entire skeletal muscle fiber. F. J. M. SICHEL. *Laboratory of Physiology, University of Vermont, College of Medicine, Burlington.*

Lengths of fibers were isolated from the adductor muscles of the frog and were mounted in a chamber for stretching between microneedles using methods previously described. Several places along the fiber were then lightly crushed between the cover glass and a smooth spherical fused microneedle tip. When such fibers were allowed to stand for 5 to 10 minutes under a slight tension the coagulated material left in the fiber by the crushing retracted lengthwise so as to leave a length of sarcolemma emptied of its fibrillar material. Such an "empty" sarcolemma is morphologically continuous with and similar to that of the intact region of the fiber. The fiber was then slowly stretched and the extensions measured in the intact regions and in the "empty" regions. In order to confine the measurements to these regions without involving those containing coagulated material, the measurements were made between suitably placed carbon particles adhering to the surface of the fiber. Since at any given length the tension in all segments of the fiber must be the same it is possible to compare at the same tensions the per cent elongation of a normal segment and of an "empty" segment.

It was found that the per cent elongation of the "empty" lengths ranged from 1.1 to 4.2 times the per cent elongation of the intact regions of the fiber, an average figure being 2.2 times. The lower range of figures for extension of the sarcolemma were obtained in preparations in which the

coagulated material did not separate cleanly from the sarcolemma. When a region included several obvious clumps of coagulated material as well as "empty" parts then the relative extensions approached very closely the simultaneous relative extensions in an intact region. It follows that the entire cross-section of the intact fiber should resist extension with a tension at least twice that due to the sarcolemma alone at a comparable extension.

Electrical anesthesia with constant currents. M. L. SILVER (by invitation) and R. W. GERARD. *Department of Physiology, University of Chicago, Chicago, Ill.*

An uninterrupted direct current passed between non-polarizable electrodes in mouth and anus of frog, rat, or dog can produce and maintain a state of immobility and non-responsiveness to stimulation. To effect this electrical anesthesia the current is increased gradually, to avoid stimulation; or, especially in dogs, the electrodes are placed and current initiated under preliminary light chemical anesthesia (evipal, 30 mgm. per kilo).

A current of 3 mA in the frog, 12 mA in the rat, and 45 mA in the dog, suffices to maintain the inactive state indefinitely—as long as 8 hours in one dog. To terminate anesthesia the current is decreased gradually or even reversed for a short time, to hasten emergence. Recovery seems complete in the frog within a minute, in the rat within 10 min., in the dog in 100 min., although its reflexes return in 15 min. Animals after as many as 8 anesthetic periods of several hours each have shown no physiological injury, nor histological changes in the spinal cord (rat and frog).

During induction of electrical anesthesia in the dog, crossed extension, flexion, and knee jerk reflexes briefly increase and then disappear in that order. Blood pressure and heart rate are not significantly altered by the anesthesia, respiration may deepen following it.

The reflex block is due to central action, since spinal root irritability is normal during electro-narcosis while motoneurone thresholds, tested with micro-electrodes, are raised. Only that current passing through the spinal cord is, therefore, effective. The resistance of dog and electrodes is nearly 500 Ohms, and some 20 volts must be applied to produce the 45 mA for anesthesia. The current in the cord averages 1.4 per cent of the total in the dog. Assuming a comparable ratio in the other animals and allowing for differences in cord cross-section, in each species a current density of about 1 mA per sq. mm. cord cross-section produces inactivity.

The distribution of current flow in body structures was measured by the IR drop between variously placed electrodes; and the influence of anesthetizing electrode positions on current flow through the cord was similarly studied. With polarizing needle electrodes in the cisterna and lumbar space spinal anesthesia results without suppressing head responses.

Acquired tolerance to small doses of post-pituitary extract.[1] H. SILVETTE and C. N. PSIMAS (by invitation). *Department of Pharmacology of the University of Virginia, Charlottesville.* (Read by title.)

[1] This investigation has been made with the assistance of a grant from the Committee on Therapeutic Research, Council on Pharmacy and Chemistry, American Medical Association.

It has been shown (Am. J. Physiol. **131**: 601, 1941) that large doses of post-pituitary solution (500 milliunits or more), injected at 2-hour intervals into diuretic rats, continuously inhibited their urine flow. It was concluded from these experiments that the renal tubule cells develop no tolerance to the extract, but are constantly sensitive to the antidiuretic hormone.

We have now tested the tolerance of white rats to small amounts of post-pituitary solution. In these experiments the dose used was 10 milliunits, and the injections were repeated at intervals of 3 days or longer.

A series of 24 animals, given 10 cc. 0.2 per cent sodium chloride solution intraperitoneally, excreted in 6 hours 7.4 cc. urine containing 0.32 mgm. chlorides per cubic centimeter (average). The same experiment repeated four days later gave identical results in urine and chloride excretion.

Another series of 24 animals was similarly treated, except that 10 milliunits of Post-Pituitary Solution (Squibb) were added to the injection fluid. These animals excreted 6.0 cc. urine containing 2.40 mgm. chlorides per cc. When this experiment was repeated four days later, however, the urine excretion rose to the normal level of 7.3 cc. in 6 hours and the chloride concentration dropped to 1.33 mgm., thus indicating an acquired tolerance to this dose of extract. Three days later, under the same experimental conditions, the animals excreted 7.1 cc. urine containing 1.54 mgm. chlorides per cc.

Following a two-weeks rest, 10 milliunits of extract given to the same animals were again found to exert a marked antidiuretic effect: urine output was reduced to 4.6 cc. while chloride concentration reached 2.33 mgm., demonstrating that the tolerance previously acquired had been lost in the fourteen-day interval. The animals were allowed to rest for sixteen days further, after which 10 milliunits of the extract still gave a similar antidiuretic response.

The tolerance to small doses of post-pituitary extract thus seems to be both quickly developed and quickly lost; and this phenomenon of tachyphylaxis indicates, we believe, an altered rate of destruction of the hormone in the extra-renal tissues rather than a change in renal tubular response.

A new precision pipette for volumetric gaseous analysis. ERNST SIMONSON (introduced by A. C. Ivy). *Mount Sinai Hospital, Milwaukee, Wis.* (Demonstration.)

The usual type of graduated pipettes for volumetric gaseous analysis (for instance in Haldane's apparatus) does not permit analysis of air mixtures with a nitrogen content less than 60 per cent. The accuracy of the Haldane 10 cc. pipette is 0.01 cc. to be read, and 0.005 cc. to be estimated. To increase the accuracy by increasing the volume has been tried. In this case, the error due to the thermobarometer and the time necessary for the analysis increases so that the 20 cc. pipette does not have double the accuracy of the 10 cc. pipette. The new system consists of 2 parallel pipettes shunted with one another for the air at the top, and for the mercury by means of a T-tube at the bottom. Each has a separate cock in the T-tube. The right pipette is subdivided into 10 bulbs each of 1 cc. The left pipette has a capacity of 1 cc. and is subdivided into 0.002 cc. Both pipettes are filled with mercury. First the right pipette

is filled with gas until the marked of the last full cc. Then the rest of the gas is filled into the 1 cc. pipette, where the final reading is taken with an accuracy five times greater than in the usual pipette of the same volume. With this system, any mixture of gases can be investigated. It may be attached to any volumetric gaseous analysis apparatus.

Investigations of the state of the central nervous system by means of the fusion frequency of flicker. Influence of age, fatigue and cerebral lesions. ERNST SIMONSON, NORBERT ENZER and SAMUEL S. BLANKSTEIN (introduced by A. H. Steinhaus). *Mount Sinai Hospital, Milwaukee, Wis.*

The fusion frequency of flicker is regarded to be a fundamental visual process. Its value depends on the state of the whole visual pathway if all experimental conditions (intensity of illumination, size of illuminated area, etc.) remain the same. The fusion frequency can be measured with great accuracy, the daily variations do not exceed 3 to 4 flashes per second when the subject's condition is not altered. There is a general definite decrease of the fusion frequency with age as demonstrated by the average and maximum values of 47 subjects between 14 and 80 years, subdivided into four age groups. The high values found in about half of the subjects between 10 and 29 years cannot be obtained in the older age groups, but on the other hand, in the young values as low as in the older age groups may be obtained. Fifty-six experiments in 19 normal subjects showed a regular decrease of the fusion frequency at the end of the working day, parallel to the amount of work done and the subjective feeling of fatigue. This indicates fatigue of the central nervous system, because muscular fatigue could be excluded in the types of occupations investigated (typing, laboratory work, secretary work, etc.). If the flash frequency is increased beyond the usual fusion frequency, provided that alternating current is used for the light source, a second flicker appears at higher frequency and after disappearing, another or a third flicker reappears at a much higher frequency. The second and third flicker is not influenced by the intensity of illumination, size of area illuminated or the relative duration of flashes. This leads to the conclusion that its perception must be due more to cerebral than to retinal processes. This is confirmed by the fact that in six patients with cerebral lesions, but intact ocular structures, the recognition of the second and third flicker was either abolished or markedly impaired.

Effects of amphetamine sulphate and antuitrin-S on gastro-intestinal motility in the rat.[1] ERMA A. SMITH. *Iowa State College, Ames.*

Doses of amphetamine sulphate ranging from 2 mgm. to 10 mgm. per rat were fed in 2 gram food pellets dyed with Fe_2O_3. Forty animals sacrificed at intervals of 30 min. to 2 hrs. following administration of the drug showed delayed rate of passage of the food along the alimentary tract. Excrementometer readings using intact animals showed delay in time of first appearance of the dye in the feces. Isolated segments of colon and duodenum contracting in Sollman's solution were inhibited by minimum effective doses and stimulated by larger doses added to the bath.

[1] Antuitrin-S was furnished by Dr. Oliver Kamm of Parke Davis and Co. Aided by a grant from the Committee on Scientific Research of the American Medical Association.

Antuitrin-S inhibited gastro-intestinal motility, even in minimum effective doses, when injected into live animals and when applied to isolated segments.

The dispersion of glomerular activity in the normal and hypertensive kidney.[1] HOMER W. SMITH, HILMERT A. RANGES (by invitation), HERBERT CHASIS (by invitation) and WILLIAM GOLDRING (by invitation). *Departments of Physiology and Medicine, New York University College of Medicine, and the Third (New York University) Medical Division of Bellevue Hospital, New York City.*

So long as the load of glucose ($p_g c_{in}$ = plasma glucose conc. × filtration rate) to a particular nephron is less than the reabsorptive capacity of the attached tubule (tm_g), glucose reabsorption is essentially complete, but as soon as the load exceeds tm_g all excess glucose is excreted in the urine. Since p_g is identical for all nephrons, one can determine the dispersion in the ratio: c_{in}/tm_g (= glomerular activity ratio), relative to the mean ratio (C_{IN}/Tm_G) for the entire kidneys, by "titrating" the kidneys with glucose and noting the value of $P_G C_{IN}/Tm_G$ at which increasing numbers of nephrons become saturated.

Repeated titrations of 10 normal, basal subjects show that the detectable limits of dispersion in c_{in}/tm_g rarely exceed the extremes of 0.66 and 1.33 of the mean. During renal ischemia elicited by orthostatic circulatory strain (tilt-table) the limits of dispersion are perceptibly widened, indicating that the filtration rate is reduced in some glomeruli more than in others, apparently by vasoconstriction on the afferent side of the glomerulus.

Some hypertensive subjects show dispersion outside the normal limits; the fact that Tm_g in these subjects may be equal to or above the mean normal value suggests that the increased dispersion is attributable to local occlusion on the afferent side and/or reduced glomerular permeability, rather than to local reduction in tm_g. In other hypertensive subjects, however, the dispersion is within the normal limits.

An attempt is being made to determine whether the increased dispersion occurs early in the course of the disease and whether, therefore, it may be considered as causal, or whether it appears only after such a period that arteriolar or glomerular lesions may have been produced by hypertensive disease.

Iron absorption in the absence of bile. PAUL W. SMITH and LATHAN A. CRANDALL, JR. *Department of Physiology, College of Medicine, University of Tennessee, Memphis.*

It has been frequently suggested that bile is necessary for the absorption of adequate amounts of iron from the intestine. Since most of the evidence for this view is of an indirect nature, we have applied a more direct method to a re-investigation of the problem.

More than 40 experiments have been completed, using 7 bile-fistula, and 12 unoperated dogs. The bile fistulae were of the internal type, in which the gall bladder is joined to the pelvis of the kidney. Ferrous sulfate or ferrous gluconate were given orally, and the inorganic iron content of the plasma was determined at intervals thereafter. Control

[1] Aided by a grant from the Commonwealth Fund.

experiments on fasting animals showed that the iron content of consecutive plasma samples taken at 2 hour intervals either remains constant or decreases. A significant rise in the iron content of the plasma was therefore considered indicative of absorption; the failure of such a rise to occur was interpreted as inhibited or delayed absorption.

The rise of the plasma iron level following the administration of ferrous sulfate in dilute solution, by stomach tube, was of approximately equal magnitude in fasting bile-fistula and unoperated animals. In the fasting, bile-fistula animal, absorption of iron from a solution of ferrous gluconate was reduced by the addition of 100 cc. of mineral oil, and was almost completely suppressed by the addition of the same quantity of olive oil. Olive oil had no effect on absorption by fasting, unoperated dogs. The addition of 1 gram of ferrous gluconate daily to the regular diet caused an approximately equal rise of the plasma iron level in bile-fistula and unoperated dogs.

Our experiments do not indicate that iron absorption from an average diet would be greatly reduced in the absence of bile.

Vocalization and other responses elicited by excitation of the Regio cingularis in the monkey. WILBUR K. SMITH. *Department of Anatomy, The University of Rochester, School of Medicine and Dentistry, Rochester, N. Y.*

Although various theories have been advanced concerning the function of the cingular region, its physiological status is unknown. The literature reveals no experimental evidence as to its probable functional rôle. In stimulation experiments on monkeys (Macaca mulatta) it has been found possible to elicit responses the sum total of which amounts to emotional expression.

Electrical stimulation of the rostral part of the cingular region (area 24 of Brodmann) under very light anesthesia produces vocalized responses identical to those which the animal makes under ordinary conditions. The one most frequently elicited consists of a low guttural sound, emitted once if the application of the stimulus is of brief duration, but repeated several times if the stimulus is applied for a longer time. In some instances a sound of higher pitch is obtained. Vocalization may occur alone, but in its fully developed form it is part of a complex act simulating emotional expression, and is characterized by opening of the eyes, dilatation of the pupils, opening of the mouth, retraction or protrusion and rounding of the lips, and vocalization. When the stimulus is applied the animal appears to awaken, when it is discontinued the eyelids close and the animal appears to fall asleep. Bilateral extirpation of the excitable area does not result in loss of the ability to vocalize.

Respiration changes markedly in character during the vocalized response, but in addition to this change, stimulation has a pronounced inhibitory effect upon respiration in the absence of vocalization. Furthermore, excitation often produces a marked inhibition of movements of the extremities, any movement in process of execution is stopped, and relaxation of the muscles ensues. A striking demonstration of the inhibitory power of this area is evidenced by the finding that the struggling which ordinarily ensues when the ether cone is applied to the lightly anesthetized animal is completely prevented or abolished by application of the stimulus just before the anesthetic is applied.

In addition to the cingular region, vocalization has been obtained from the region of the uncus, from the hypothalamus by excitation of the wall of the third ventricle, and from the mesencephalon.

Inhibition of bladder contraction by the cerebral cortex. WILBUR K. SMITH and GEORGE C. WHITNEY (by invitation). *Department of Anatomy, The University of Rochester, School of Medicine and Dentistry, Rochester, N. Y.* (Read by title.)

Electrical excitation of a cortical area on the orbital surface of the frontal lobe in the cat has previously been shown to produce marked inhibition of respiratory movements and rise of arterial pressure (Smith, 1938), and decrease of tonus in the gastric musculature (Bailey and Sweet, 1940). Further study of the responses of this area shows that it is able to exert a powerful inhibitory effect upon the bladder.

Experiments were performed on adult non-pregnant female cats lightly anesthetized with ether. Intravesical pressure was measured by means of a straight glass tube manometer connected to a catheter inserted into the bladder through the urethra, the catheter being sufficiently small to permit voiding during relaxation of the sphincters. Warm Ringer's solution was used to distend the bladder to a degree sufficient to cause spontaneous contractions. Kymographic records were made by connecting the open end of the manometer to a Marey tambour.

Spontaneous micturition under these conditions usually is preceded by small rises of pressure gradually increasing in amplitude and frequency over a short period. These are followed by an abrupt elevation of pressure rapidly attaining a peak, which is sustained for some seconds during which fluid begins to escape. The sustained rise is followed by a further loss of fluid and fall in pressure usually occurring in steps over a period of several seconds.

Stimulation of the inhibitory area during the initial small rises of pressure leads to their temporary abolition. Application of the stimulus during the main rising phase preceding the relaxation of the sphincters usually causes the pressure to return to or below the former resting level. Stimulation applied after the peak pressure has been attained markedly shortens the duration of the sustained rise. If stimulation is continued, the decline in pressure without the escape of fluid is rapid, and the step-like character of the decline is absent, indicating inhibition of the stretch reflex. Under these conditions the pressure may fall below the preceding resting level. Simultaneous recording of respiratory movements and intravesical pressure indicates that alterations in the two are independent.

Glomerular filtration and renal blood flow in the rabbit. WILLIE W. SMITH (introduced by Homer W. Smith). *Department of Zoology, Smith College, Northampton, Mass., and Mt. Desert Island Biological Laboratory, Salsbury Cove, Me.*

Following methods developed by H. W. Smith and associates, rates of glomerular filtration and renal blood flow were simultaneously determined for rabbits under basal conditions and with mannitol diuresis, and relationships which would result from changes in afferent resistance alone, or in efferent resistance alone, were derived.

In basal experiments no correlation was apparent between filtration

rate and urine formation. This is not in agreement with the data of Kaplan and Smith, but a reexamination of their experiments leads us to suggest that the low filtration rates for low urine volumes might have been due to effects of excessive hydration. In mannitol diuresis the filtration rate averages 38 per cent above the mean control figure, but within the group there is no correlation between filtration rate and urine formation.

Individual rabbits vary in the relationship between filtration rate and blood flow. In general, an increase in blood flow is accompanied by an increase in filtration rate, the filtration fraction remaining nearly constant. This indicates that both afferent and efferent arterioles participate in the control of the glomerular circulation, afferent contribution being more marked at relatively low blood flow. In mannitol diuresis both blood flow and filtration rate are increased but the filtration fraction tends to fall.

One animal had previously undergone unilateral nephrectomy, which resulted in hypertrophy of the remaining kidney; in this rabbit the relationships between filtration rate and blood flow were like those of a normal rabbit.

Attempts to induce hyperemia by the use of pyrogenic inulin were unsuccessful.

The enterohepatic circulation of bile pigment. E. F. SNAPP and A. L. BERMAN (introduced by K. K. Jones). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Investigation of this problem was undertaken because of the conflicting evidence regarding the existence of an enterohepatic circulation of bile pigment.

Chronic bile fistula dogs of the Rous-McMaster and the suction-biliary duodenal type were used to study the absorption of purified bilirubin from the intestine, the normal variations in pigment output, and the effects of bile salt preparations and whole dog bile upon pigment output. In acute experiments we studied the lymphatic absorption of bilirubin from the intestine.

The dogs were fed a standard measured diet, and except for control periods of 3 to 5 days were maintained on bile salt preparations or whole bile given at each meal time. The bile salts preparations which were given in 3 or 5 gm. doses daily were ox-bile salts and oxidized bile acids. Whole bile was returned to the dogs on suction through a tube into the duodenum.

The average daily pigment output was 9.7 mgm. per kilo. body weight. The daily output varied as much as 40 per cent from the mean in the dogs on bag collection and as much as 25 per cent in the dogs on suction. When bile salt preparations were administered there was no significant increase in the total pigment output. When whole dog bile was returned into the intestine, the average daily pigment output was increased 40 per cent. When commercial preparations of pure bilirubin were given to chronic biliary fistula dogs, there was no increase in the daily total pigment and bilirubin output. In 8 experiments, when pure bilirubin plus gall bladder bile was placed in the duodenum, only minute quantities could be found in the thoracic duct lymph.

Thus, commercial preparations of pure bilirubin were not absorbed, whereas when whole bile was given the pigment output was significantly

increased. This is probably due to the difference between the chemical and physical properties of the pure bilirubin and the bilirubin as it occurs in bile.

Extensor rigidity in cats produced by simultaneous ablation of the anterior lobe of the cerebellum and the pericruciate areas of the cerebral hemispheres.[1] RAY S. SNIDER (by invitation) and CLINTON N. WOOLSEY. *The Johns Hopkins University, School of Medicine, Baltimore, Md.*

Since Sherrington's (1896) description of decerebrate rigidity, it has been known that electrical stimulation of the anterior lobe of the cerebellum inhibits decerebrate posture. Bremer (1922) and Bremer and Ley (1927) showed that ablation of the anterior cerebellar lobe produces increased extensor tonus and increased positive supporting reactions in normal cats and pigeons, and an augmentation of rigidity in previously decerebrated cats. Fulton and Connor (1939) found that ablation of the anterior lobe, exclusive of the lingula, causes in dogs and monkeys "increased tendon reflexes, pronounced lengthening and shortening reactions and gross exaggeration of positive supporting reactions." Studies on the role of the cerebral cortex in the control of posture have demonstrated that bilateral ablation of pericruciate cortex in carnivores produces extensor rigidity, comparable in degree with that of decerebrates. It is best seen when the preparation is suspended. (See Bard, *Macleod's Physiology*, 1941, p. 136.)

We have simultaneously ablated the anterior lobe of the cerebellum and the pericruciate areas of both cerebral hemispheres of the cat and have obtained a preparation in which apparently the separate effects of the cerebral and the cerebellar ablations are summated to produce an intensely exaggerated antigravity attitude. The result is a pillar-like rigidity of all four limbs, to which extensors, flexors, abductors and adductors contribute. A similar increase of activity occurs also in the dorsal and ventral muscles of neck, trunk and tail. The effect of the antigravity muscles exceeds that of the flexors and the result is an exaggerated caricature of standing with fore- and hindlimbs rigidly extended, elevation and retraction of the head, arching of the back and elevation of the tail. The attitude may persist with little diminution in intensity for several days, (acute sterile preparations). Muscles stand out in relief beneath the skin and exhibit such marked enhancement of stretch reflexes that reflex contractions may be elicited easily by tapping the muscles.

The preparation is being used in a study relating muscular activity to muscular atrophy.

Teledeltos paper polygraph. J. M. SNODGRASS (introduced by Charles G. Rogers). *Oscillograph Laboratory, Department of Psychology, Oberlin College, Oberlin, O.* (Demonstration.)

The Teledeltos polygraph gives a jet black, permanent record without the use of ink. Any device capable of recording on smoked paper will operate satisfactorily. The "Teledeltos" paper will record from an almost imperceptible speed to a speed high enough to resolve a 10,000 cycle wave.

A glow lamp record and demonstration of muscle function in movement. J. M. SNODGRASS and G. F. MAHL (introduced by Charles G. Rogers). *Oscillograph Laboratory, Department of Psychology, Oberlin College,*

[1] Sponsored by The National Foundation for Infantile Paralysis.

Oberlin, O. (Motion picture demonstration.)

The action potential output from cutaneous pad electrodes is amplified; and the amplifier output connected to neon glow lamps placed on the muscle or moving member. The electrical phase of the contraction lights its neon lamp. This is a convenient application of action current recording for the study of posture and phasic movements. The technic is applicable in checking after therapy of poliomyelitis cases, and other forms of muscular insufficiency.

Mammalian muscle action potentials of less than a millisecond. J. M. SNODGRASS and R. W. SPERRY (introduced by Hallowell Davis). *Oscillograph Laboratory, Department of Psychology, Oberlin College, Oberlin, O.*

Muscle action-potential pulses having time relations approximating the discharges in mammalian motor nerve fibers have been obtained from human subjects. The potentials may be picked up from the extensor communis digitorum by either concentric needle electrodes or cutaneous pad electrodes. The potentials are characterized by a "spike" potential of approximately 0.4 msec. duration, followed by a relatively slow "negative after-potential" lasting from 1.5 to 3 msec. For detailed analysis the action potentials are recorded optically as a sound track on 35 mm. sound film. The recording is transcribed using sound-film speeds from the recording speed to 1/100 of the recording speed. A photo-electric pickup is used in transcribing. A direct-coupled amplifier connected to the photo-electric pickup operates a Westinghouse oscillograph. At a foot and a half per second, a sound film resolves 20 impulses per millisecond. This "slow transcription system" extends a millisecond to 5 cm. or more with high precision. Both the time-axis and the amplitude may be independently expanded. This same technic can be used with an instantaneous disc recording. By this flexible technic the summation of the action-potential pattern has been closely followed.

Differences of rates of flow through portal and venous systems of the liver under various conditions.[1] CHARLES D. SNYDER and FRANK H. TYLER (by invitation). *Department of Physiology, Johns Hopkins University, School of Medicine, Baltimore, Md.* (Read by title.)

The hepatic venous musculature of *Pseudemys elegans* as shown by Tyler (The Anat. Rec. 1941, in press) has a "sluice-mechanism" resembling that of the dog. The walls of portal venules and sinusoids are thin and devoid of muscle, while few, widely scattered muscle cells are found in the walls of the portal vein itself. The lobular arrangement of parenchyma, biliary and arterial walls resemble human liver.

Observations of simultaneous flow-rates through portal and hepatic paths under a variety of conditions have been made. All findings are reduced to standard terms of volume flow per unit time per unit weight of organ.

A demonstration of differences in effects of vagal and splanchnic nerve stimulation, or of autonomomimetic drugs, upon initial responses of both inflow and outflow rates, (1) in latencies of initial response, (2) in time to maximum of initial response should yield answers to questions as to which of the venous systems are affected and as to the nature of the effects.

In response to acetyl choline, mean duration to midpoint of maximal

[1] Aided by a grant from the Rockefeller Foundation Fluid Research Fund.

initial response is twice as long in the inflow rate as it is in the outflow rate. This indicates the effect is due to the hepatic venules which, constricting peristaltic-wise from central to peripheral regions, produces brief, enormous increase in hepatic outflow. This constriction sets up immediate resistance to inflow. During hepatic vein constriction, inflow head of pressure remaining constant, dilatation of sinusoids and portal veins (whose walls are thin and provided with elastic tissue fibers) follows. Decrease in inflow, therefore remains less than increase in outflow. When hepatic veins relax, this accumulating portal fluid suddenly finds outlet; rate of hepatic outflow increases rapidly or slowly, according to dose of drug, until rates of flow approach again initial levels. Out of 9 livers studied one only did not follow this pattern of behavior as just described.

The effects of epinephrine on vascular response are likewise analyzed; their nature is not so clear, but will be discussed, as will also the latencies of initial responses.

A comparison of the effects of upper and lower motor neurone lesions on skeletal muscle.[1] D. Y. Solandt and J. W. Magladery (by invitation). *Departments of Physiology and of Physiological Hygiene, University of Toronto, Toronto, Canada.*

Experiments were performed on albino rats in which denervated gastrocnemius muscles were compared with those deprived of their upper motor neurone control by spinal cord section. In the majority of the experiments the cord was sectioned about the level of the 6th thoracic vertebra and the sciatic nerve on one side was cut. The denervated muscles showed the typical fibrillation while those deprived of upper motor neurone control showed hyperexcitability, some spasticity, but no activity resembling fibrillation. For the first two weeks the atrophy in the two types of muscles was of approximately the same degree. In both cases the muscles were reduced to about half their calculated normal weight at the end of this time. When left for longer periods the denervated muscles continued to atrophy while the spastic muscles started to regain weight.

Two weeks after sciatic nerve section the denervated muscles were found to react to less than one one-millionth the quantity of intra-arterially injected acetylcholine which would just produce a response in normal muscle. Similarly tested two weeks after cord section the spastic muscles responded to only one one-hundredth the dose of acetylcholine which was effective for normal muscles. In these latter muscles the sensitivity to acetylcholine tended to return towards normal as the muscles regained weight.

The activated collodion membrane and its electrochemical behavior. Karl Sollner, Irving Abrams and Charles W. Carr (introduced by M. B. Visscher). *Department of Physiology, University of Minnesota, Minneapolis.*

It was reported briefly (Sollner and Abrams, J. Gen. Physiol. **24:** 1, 1940) that collodion membranes prepared from pure collodion do not show the characteristic electrical phenomena reported by previous authors. We could not duplicate, for example, the high concentration potentials

[1] Sponsored by the National Foundation for Infantile Paralysis, Inc.

with dried membranes (Michaelis) and strong anomalous osmosis with porous membranes (Loeb, Bartell). It was shown (Sollner, Abrams and Carr, J. Gen. Physiol., **24**: 467,1941) that the electrochemical behavior of collodion is caused substantially by partial oxidation, specific ion adsorption playing only a minor role. The charge density at the collodion surfaces, determined by the number of COOH-groups of the different preparations, is the ultimately decisive factor. This charge density may be vastly increased by proper oxidation. Suitable oxidizing agents are sodium and calcium hypochlorite and particularly sodium hypobromite. In addition, collodion may be activated by treatment with strong alkalies, because it undergoes progressive decomposition of an oxidative nature in contact with alkaline solutions.

The behavior of the membranes towards nonelectrolyte solutions is practically identical before and after oxidation. We have reason to believe, therefore, that their spacial structure is hardly affected by oxidation. Their behavior towards electrolytes is changed completely by oxidation. With dried membranes the concentration potential reaches the thermodynamically possible maximum and anomalous osmosis is extremely marked with the porous membranes.

The Meyer and Sievers theory of electrochemical membrane behavior (Helv. Chim. Acta **19**: 649, 665, 1935), predicts appreciable base exchange capacities for membranes having medium or high electrochemical activity. The experimental values obtained by careful analytical and electrometrical determinations are lower by several orders of magnitude than those calculated according to the Meyer and Sievers theory. In the authors' opinion, the irregular shape of the pores could readily account for this discrepancy. The electrochemical properties of membranes are determined by the properties of individual spots within the pores, rather than by (calculated) average values.

The relation between the phosphate changes in blood and muscle, following dextrose, insulin and epinephrin administration. SAMUEL SOSKIN, R. LEVINE (by invitation) and O. HECHTER (by invitation). *Department of Metabolism and Endocrinology, Michael Reese Hospital, and the Department of Physiology, University of Chicago, Chicago, Ill.*

The administration of dextrose, insulin and epinephrin respectively to normal animals causes a fall in the inorganic phosphate of the blood. Insulin and epinephrin also result in a coincident rise in the hexose monophosphate content of the muscle. It has been assumed that the disappearing blood phosphate enters the muscle together with blood glucose.

A comparison of the effects obtained in normal, depancreatized and adrenalectomized animals has revealed the fact that the blood and muscle phosphate changes are not directly related to each other. The fall in blood inorganic phosphate is not reflected in the total phosphate content of the blood, and is probably due to an esterification within the red blood cells. Insulin decreases the blood inorganic phosphate under all circumstances, while dextrose and epinephrin fail to do so unless insulin is present. On the other hand, epinephrin increases the hexose monophosphate content of muscle under all circumstances, while insulin fails to do so unless epinephrin is present. The usual observation of both blood and muscle effects with insulin and epinephrin in the intact normal animal, is due to

the reflex evocation of the secretion of one gland by the effects of the hormone of the other.

The action of insulin in esterifying blood inorganic phosphate, is considered in relation to the general action of insulin on carbohydrate metabolism.

The initial stabilization period of the perfused frog heart. C. R. SPEALMAN. *Department of Physiology and Pharmacology, Medical College of Virginia, Richmond.*

During the first period of perfusion, the rate of beat of the frog heart gradually decreases and finally reaches a stable level. The amplitude of contraction also decreases during this period; but this effect may be masked partially because of the changes in rate.

I have assumed that the above effects may be due to the liberation of sympathin from sympathetic nerve endings stimulated by the sudden change in environment from blood to Ringer's solution. The experimental results described below support this hypothesis. *Experiments to determine if an active substance is liberated during the stabilization period.* These were so designed that the perfusate of a heart during this period could be passed directly through a second (stabilized) heart, with minimal delay and no disturbance to the perfusion of the second heart. In 8 experiments, the amplitude of contraction of the second heart was increased 15 to 70 per cent by the perfusate from the first heart during its stabilization period. The rate of beat of the second heart was occasionally markedly increased. Three control experiments showed that no active substance is liberated by a stabilized heart. The active substance liberated during the period of stabilization loses its activity rather rapidly. In 3 experiments, the perfusate was inactive after 45 minutes. *Experiments designed to prevent the action of any liberated sympathin.* Frogs (4) were injected with 1 cc. of a 0.1 per cent solution of ergotoxine ethanesulfonate in alcohol. Control frogs (4) received 1 cc. of alcohol. The following table gives the average of the heart rate values in beats per minute at 15 minute intervals from the beginning of perfusion for these experimental and control hearts.

	TIME IN MINUTES				
	0	15	30	45	60
Average heart rate (exp.).....	40	40	39	37	36
Average heart rate (con.).....	58	51	45	41	40

The table indicates that the typical period of stabilization is prevented by ergotoxine. It was also shown with 3 hearts that this treatment with ergotoxine temporarily abolishes or greatly diminishes the response of the heart to epinephrine.

Convulsive reactivity in hypercholesteremia.[1] E. SPIEGEL and H. WYCIS (by invitation). *Department of Experimental Neurology, D. J. McCarthy Foundation, Temple University, School of Medicine, Philadelphia, Pa.* (Read by title.)

[1] Aided by a grant of the American Medical Association, Committee on Scientific Research to E. S.

In view of experimental and clinical reports indicating a possible importance of cholesterol for the convulsive reactivity, hypercholesteremia was produced in rabbits by oral application of 1.5 grams cholesterol 3 times a week, by intraperitoneal injection of a 3 per cent emulsion in olive oil (3–5 daily injections of 5–7 cc. per kilo body weight), or by intravenous injection of 1.5 per cent colloidal solution of cholesterol in water. The convulsive reactivity was determined by electric stimulation with the skull intact as previously described (Spiegel. J. Lab. and Clin. Med. **22:** 1274, 1937). A definite change of the threshold failed to appear in all these experiments even if the level of the serum cholesterol was raised as high as 875 mgm. per 100 cc.; the changes in the duration of the convulsions were also insignificant.

Quantitative relationship between polarizability and permeability.[1] M. Spiegel-Adolf (by invitation) and E. Spiegel. *Departments of Colloid Chemistry and of Experimental Neurology, D. J. McCarthy Foundation, Temple University School of Medicine, Philadelphia, Pa.* (Read by title.)

In former experiments a method for studying the polarizability of tissues was described and used on the brain (J. Nerv. Ment. Dis. **90:** 188, 1939). Studies on artificial membranes (Spiegel-Adolf. J. Gen. Physiol. **20:** 695, 1937) indicated that polarizability is related to permeability; a further quantitative study of this relationship on a simple biological object seemed, however, desirable. On preparations from the skins of winter frogs studies of the electrolyte permeability (dialysis of N NaCl from the outer surface against 5.4 per cent dextrose solution on the inner surface) and of the polarizability were made. The polarization index Δ was determined before and after each dialysis experiment ($\Delta = 100 (C_h - C_l)/C_l$; C_h = conductivity across the membrane in Ringer solution at 5120 cycles; C_l = conductivity at 547 cycles).

A series of such successive measurements of permeability and polarizability was performed during the gradual deterioration of the skin, and the permeability values (milligrams of NaCl dialysed in 50 minute intervals as determined by conductivity measurements) were plotted against the corresponding mean Δ values. The resultant graph showed a straight line relationship between these two values. In a similar way a relationship between the permeability for a non-electrolyte (urea plus dextrose against dextrose) and Δ could be demonstrated using interferometry for determination of the amount of urea dialysed. Furthermore the polarization index of the skin could be determined by placing platinum-Ringer electrodes (Pt wire immersed in a glass tube filled with Ringer solution and closed by a cellophane membrane) on its outer surface, and it could be demonstrated that decrease of the Δ values obtained by this method is also associated with increase of the electrolyte permeability of the skin. These experiments seem to give further evidence that changes in permeability of cellular surface films may be indirectly determined by measurement of the polarization index.

Further studies on the effects of chemical inhibitors on the respiration of resting and caffeinized frog muscle. J. N. Stannard. *School of Medicine, Emory University, Ga.* (Read by title.)

[1] Aided by a grant from the American Medical Association, Committee on Scientific Research to E. S.

In an earlier publication (Am. J. Physiol. **126**: 196, 1939) it was proposed, on the basis of differential sensitivity to azide and cyanide, that the resting and "activity" respirations of frog muscle were qualitatively distinct, and that the cytochrome-cytochrome oxidase system functioned only after electrical or chemical stimulation. The present study is concerned with the action of several additional chemical inhibitors on O_2 uptake of resting muscle and muscle whose respiration was stimulated by caffeine in sub-contracture doses. The results in general are consistent with the qualitative fractionation proposed earlier.

Sodium arsenite had no effect at 0.002 M on resting respiration until the third hour. The increase in respiration induced by caffeine was largely eliminated within two hours if arsenite was added to caffeinized muscle, or prevented if caffeine was added to arsenite-treated muscle. At 0.01–0.02 M the caffeine increment was eliminated in one hour; the resting respiration decreased about 20 *per cent.*

With NaF, at 0.01 to 0.17 M, the resting respiration was either increased slightly or not affected for two hours while the extra O_2 uptake due to caffeine was reduced to zero within this interval. Fluoride-treated muscles did not respond to added-caffeine. Similar results were obtained with hydroxylamine.

Sodium pyrophosphate (0.02 0.04 M) did not interfere with either fraction of the respiration.

The "copper inhibitors," thiourea, diethyl dithiocarbamate, and potassium ethyl xanthate, either increased or caused no significant alteration in the resting respiration. At relatively high concentrations (0.01–0.03 M) they brought about complete inhibition of the caffeine increment in about three hours. These concentrations approximate those used by Graubard on uterine muscle and his enzyme preparation, but are considerably higher than those necessary to inhibit some other copper catalyses. In view of the high concentrations and length of time required for action judgement is withheld as to whether or not this represents specific inhibition of a copper-containing enzyme in frog muscle.

With the inhibitors thus far employed continuation of the resting respiration was not dependent upon the maintenance of irritability.

The relation between production of electrical energy and the oxygen consumption in surviving frog skin. PAUL STAPP (introduced by Ancel Keys). *Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis.*

By means of an iodine coulometer of low electrical resistance and minimum polarization at current densities measured, the quantity of current in the external circuit of surviving frog skin was measured for intervals of 30 minutes to three hours. Simultaneous determinations of potential and of oxygen consumption by the Winkler technique were carried out. From the ratio of calorie equivalent of joules of electricity in the external circuit to calorie equivalent of oxygen consumed for a given respiratory quotient, it was possible to determine the efficiency of electrical energy production in the external circuit of the skin. Forty-seven determinations totalling 120 hours gave values averaging 1.3 per cent for one and two hour determinations and 2.3 per cent for half hour determinations. The efficiency of total electrical energy production is

in the order of magnitude of 5 or 6 per cent. Further evidences for a relationship between respiratory processes and electrical energy production are presented by the effect of varying oxygen tension and by the accumulation of electrical energy during 8 to 10 hours of exposure to saturated air.

The effect of insulin on the respiration of human diabetic muscle. F. J. STARE (by invitation) and H. T. RICKETTS. *The School of Medicine and the Department of Medicine, University of Chicago, Chicago, Ill.*

The respiration of minced human muscle obtained by biopsy of the gastrocnemius under local (subcutaneous) anesthesia was studied in the Warburg apparatus in a glucose-Ringer-phosphate buffer at a pH of 7.4 and temperature of 38°C., with and without the addition of insulin. The tissue was minced in a special micro-Latapie apparatus. The subjects used were: severely diabetic patients who were normally sensitive to insulin clinically; diabetic patients who were resistant to insulin; and 2 acromegalic diabetics who required large doses of insulin. For control experiments, samples of skeletal muscle were taken from individuals without metabolic disease who were operated upon for other reasons.

The Q_{O_2} of the unsupplemented muscle from most diabetic patients did not differ from that of normal muscle. In the tissue from the "insulin sensitive" diabetics, insulin alone, or in one case only after the addition of fumarate, caused an increase of about 40 per cent in the respiration. This insulin effect was not obtained in muscle from the "insulin resistant" diabetics, from the acromegalic diabetics or from normal persons. Fumarate increased the respiration of normal and diabetic muscle but not that of muscle from the one of the patients with acromegaly and diabetes.

When 0.02 M pyruvate was substituted for glucose in the buffer, no effect of insulin on the oxygen uptake was observed. Fumarate, however, markedly increased muscle respiration in the presence of pyruvate.

The vertical ballistocardiograph; changes in the cardiac output on assuming the erect posture, with a further theoretical study of the blood's impacts. ISAAC STARR and A. J. RAWSON (by invitation). *Department of Research Therapeutics, University of Pennsylvania, Philadelphia.*

The impacts of the circulating blood have been recalculated by a method taking better account of vessel curvature and other anatomical features. The results are much like our first approximation, i.e., the shape of the ballistic record is accounted for and the size also, within the limits of accuracy of the assumptions.

A second ballistocardiograph has been constructed; the vertical movements of a light platform, suspended from a rigid frame by flat steel springs, are magnified and photographed. Adjusted to the same calibration as the horizontal instrument (Am. J. Physiol. **127**: 1, 139) its vibration frequency is faster when both are weighted with iron.

To avoid serious interference from vibrations in the building, the vertical instrument is mounted on steel plates weighing 480 lbs. supported on one edge by rubber-like material, on the opposite edge by a row of tennis balls.

The records obtained from normal subjects standing are practically identical with horizontal ballistocardiograms but they are more subject to

distortion from interfering vibrations, especially those arising from muscular tremors. Weak patients usually tremble sufficiently to ruin the record.

Subjects have been tested after 15 minutes rest horizontal, and then 1 and $2\frac{1}{2}$, or $2\frac{1}{2}$ and 5 minutes after arising, the duplicates agreeing well in most subjects.

Three trained subjects were tested repeatedly and the vertical cardiac outputs per minute averaged 84, 93 and 84 per cent of their horizontal values. The first subject was tested on 9 days at intervals throughout the year, the $\frac{V}{H}$ ratio ranging from 88 to 79 per cent, the low value occurring in July. In the other subjects, tested less frequently, the ratio ranged from 108 to 80 per cent and 100 to 81 per cent respectively.

In twenty three medical students tested for the first time during class exercises, the average standing circulation was identical with that lying.

In certain patients, often those with symptoms on assuming the erect position, the $\frac{V}{H}$ ratio exceeded anything encountered in healthy persons, and this abnormality could sometimes be corrected by an abdominal binder. The drug paredrine produced supernormal $\frac{V}{H}$ ratios in each of 4 healthy subjects.

Observations on the rate of volume change of the colon following the administration of magnesium sulphate and fluid enemas. F. R. Steggerda and H. E. Essex. *Department of Physiology, University of Illinois, Urbana, and The Mayo Foundation, Rochester, Minn.*

Studies were made on dogs, whose colons had previously been made opaque to x-rays by injecting thorotrast into the walls. X-ray plates were taken and volume changes calculated of the colon following the administration of magnesium sulphate and water enemas.

Dogs without food for 18 hours showed a marked increase in colon size following the administration of magnesium sulphate ($\frac{1}{2}$ gram per kilo) by stomach tube. The measured volume change showed that the colon increased in size approximately 200 per cent before defecation took place $5\frac{1}{2}$ hours after the administration of the cathartic. If the animals were fed 4 hours previous to the administration of similar doses of magnesium sulphate, defecation took place in $2\frac{1}{2}$ hours with a colon size increase of 100 per cent.

That the major effect of the colon distention is related to the emptying of the small intestine is shown by the observation that when similar doses of magnesium sulphate are given by enema, less than a 50 per cent increase in colon size is recorded with no defecation.

Studies on the rate of absorption of water from the colon were also made, by taking a series of pictures of colon size following the administration of known volumes of water into the empty colon. The absorption of water was found to be at a uniform rate with the average absorption calculated to be 78 cc. per hour.

Modification of pancreatic response to secretin by urine and urine concentrates.[1] Irving F. Stein, Jr. (by invitation) and Harry Greengard.

[1] Aided in part by a grant from the Josiah Macy, Jr. Foundation.

Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.

Following the demonstration of a secretin-inactivating enzyme in blood serum, attempts were made to identify this principle in the urine. When samples of untreated and of dialyzed normal urine were incubated with secretin, an inactivation of the secretin occurred; and such inactivation did not take place when the urine was previously boiled and cooled. The findings were similar in the case of dialyzed urine concentrated by vacuum distillation and acetone precipitation, and with urogastrone concentrates prepared by the method of Gray and co-workers. However, these concentrates were also found to decrease the responsiveness of the pancreas to secretin for some time after their injection; they also caused a decrease in the continuous pancreatic secretion elicited by injection of secretin at a constant rate. This was the case in both unboiled and boiled preparations. Apparently the urine contains two factors—a thermolabile one which acts on secretin and may be identical with the secretinase detected in blood serum, and a thermostable one which directly affects the secretory activity of the pancreas.

Observations on the dynamics of experimental pulmonary embolism.[1]

F. S. Steinitz (by invitation), R. S. Megibow (by invitation) and L. N. Katz. *Cardiovascular Department, Michael Reese Hospital, Chicago, Ill.*

The mechanism of death and the circulatory changes following major and multiple minor experimentally produced pulmonary embolism have been investigated in 14 normal anesthetized or trained unanesthetized dogs by directly measuring the pulmonary and the systemic arterial blood pressures with Hamilton manometers, the systemic venous pressure with a water manometer, the heart rate and the respiratory rate. A modification of the London cannula was employed to obtain the pulmonary arterial pressure.

These studies indicate that major and multiple minor pulmonary embolism is followed by the abrupt and persistent development of systolic and diastolic pulmonary hypertension. The degree of hypertension appears to depend upon the extent of mechanical obstruction to the blood flow through the lesser circuit. Systemic arterial blood pressure changes are not striking. The systemic venous pressure most frequently rises, rarely it falls or shows no change. Alterations in cardiac rate are inconstant. Ventricular fibrillation appears terminally in an occasional animal and may account for a number of clinical cases of so-termed "sudden death," following pulmonary embolism. Rapid respiration develops usually coincidentally with or soon after the appearance of pulmonary hypertension and tends to increase progressively. Usually, progressive cyanosis accompanies the respiratory changes.

These experiments lend little support to the theory of reflex coronary vasoconstriction as a cause of death, since mechanical factors are in themselves adequate to explain the possible changes in coronary circulation.

The cause of death in experimental pulmonary embolism appears to be due to a rapid or a gradual failure of the right heart, brought about by the pulmonary arterial or diffuse arteriolar obstruction; so called hypokinetic circulatory failure being in reality rapid right heart failure.

[1] Aided by the A. D. Nast Fund for Cardiac Research.

The effect of narcotics on the resting and activity oxygen consumption of frog muscle. JOSEPH R. STERN (by invitation) and KENNETH C. FISHER. *Department of Biology, University of Toronto, Toronto, Canada.*

From the quantitative effects of ethyl on the respiration of several types of cell it is possible to show that the total respiration is composed of two discrete parts (Fisher et al., abstracts in Biol. Bull. Oct., 1940).

The "activity" of cell division in yeast and in fertilized sea urchin eggs, and the production of light by luminous bacteria, appear to be associated with only one of the two systems. In view of the similar separation of a resting from an activity oxygen consumption in frog muscle from the effects of azide (Stannard. Am. J. Physiol. **126:** 196, 1939) it was of interest to examine the effects of narcotics on muscle respiration.

Intact frog muscles were prepared as described by Stannard. After establishment of the control rate of respiration (resting or caffeinized) the narcotic was added from the onset. Ethyl urethane did not inhibit the resting oxygen consumption except at high concentrations. Sucrose, at an equivalent molar concentration produces a very similar effect. Marked inhibition of the activity oxygen consumption by urethane was however observed to occur with concentrations which had little (acceleration) or no effect on the resting oxygen consumption.

Chloretone and luminal did not inhibit the respiration of winter muscle but brought the respiration of the caffeinized muscle down to the range of the resting rates.

Muscles from spring frogs were not used by Stannard. These muscles were found to have resting rates of 40–60 cu. mm./gm./hr. Chloretone inhibited both the resting and activity respirations of these muscles but the mass law analysis shows that the two respirations are qualitatively distinct.

The resting respiration of winter muscle appears to be narcotic insensitive. In spring muscle both resting and "activity" respirations are narcotic sensitive. Narcotics appear to identify an "activity" metabolism whether that is obviously superimposed upon a resting one, as in muscle, or whether the two are incorporated into a fixed total, as in yeast.

Evidence for a neural quantum in sensory discrimination. S. S. STEVENS. C. T. MORGAN (by invitation) and J. VOLKMANN (by invitation). *Department of Psychology, Harvard University, Cambridge, Mass.*

A quantal theory of the neural processes mediating sensory discrimination has been formulated and subjected to experimental test.

The substance of the quantal theory is this: 1, a given sensory stimulus produces an all-or-none excitation of some number of neural units (quanta) and there is left a small surplus of stimulation subliminal to the excitation of one additional unit; 2, the number of units excited, as well as the amount of subliminal stimulation of the additional unit, fluctuates randomly during the presentation of a constant stimulus.

Under experimental conditions in which human observers are required to react to an abrupt increment in the frequency (or the intensity) of a pure tone, the quantal theory predicts 1, a rectilinear relation between the size of the increment and the relative frequency with which it will be detected, and 2, that the slope of this rectilinear function will be such that the increment perceived 100 per cent of the time is twice the largest increment which is never detected.

Experiments with increments of intensity and of frequency gave results conforming to these predictions. Data from these experiments provide a precise measure of the size of the neural quantum. This was found to vary with individuals and to be related to the intensity of the stimulus.

The non-peripheral locus of the neural quantum was demonstrated by measuring the frequency-quantum binaurally and monaurally. The predictions of the quantal theory were satisfied equally well in both cases, but in most observers the binaural quantum was about two-thirds the size of the monaural quantum. Apparently, therefore, the quantum can not be identified with a single afferent neuron, but is probably centrally located. That it is a *functional* rather than an *anatomic* unit seems probable.

A cyanide-substrain of yeast for studies of *in vivo* chemical organization. THEODORE J. B. STIER and JOHN G. B. CASTOR (by invitation). *Biological Laboratories, Harvard University, Cambridge, Mass.* (Read by title.)

A method of circumventing many of the experimental difficulties and limitations encountered in using "specific" inhibitors for studies of *in vivo* chemical organization has been on trial in this laboratory for several years. It consists of treating a parent strain of yeast during cell proliferation with an enzyme poison until a pure substrain which will maintain its new altered characteristics in the absence of the modifying agent is obtained. Our first attempt with KCN as the modifying agent has given a pure substrain (isolated from a single cell) which exhibits metabolic properties generally characteristic of cyanide-poisoned yeast cells. Thus, it consumes 85 per cent less oxygen and produces about 25 per cent more carbon dioxide in dextrose-phosphate solutions than the parent strain; its respiration is cyanide and azide-insensitive; "indophenol oxidase" is absent; glucose dehydrogenase and catalase activities are the same in both strains. These properties have been exhibited with great constancy during the past five years.

We can thus make available at any time unlimited quantities of cells possessing the same altered physiological characteristics and, presumably, the same altered *in vivo* chemical organization. Especially important is the possibility of analyzing these cells directly by chemical methods or indirectly by physiological methods for their component enzyme systems without interference from the chemical agent originally employed in modifying the cells.

It is our plan, as time permits, to attempt the production of additional substrains of the parent strain of yeast by use of other enzyme poisons and various physical agents which bring about modifications of cellular metabolism. It is hoped that by this program we will be able to delete (or modify) various constitutive enzymes and thus ascertain the probable *in vivo* rôle of each deleted unit in the chemical organization of the "normal" parent cell.

The relative effects of aluminum hydroxide and aluminum sulfate on the absorption of dietary phosphorus by the rat. HAROLD R. STREET (by invitation) and O. W. BARLOW. *Research Laboratories of the Winthrop Chemical Company, Inc., Rensselaer, N. Y.*

The prolonged clinical use of antacids may give rise to more or less definite adverse effects on digestion and the normal acid base balance. The

increasing clinical use of aluminum hydroxide, which in certain respects differs from usual fixed alkalies in its antacid effects, nevertheless raises the question as to the importance of possible deleterious effects of aluminum and of the basic salts in particular on the phosphorus balance.

The growth rates of young rats on the low phosphorus diet of Schneider and Steenbock have been shown to be proportional to the percentage of available phosphorus incorporated in the diet. Under such conditions a normal rate of growth was observed when the ration contained 0.24 per cent of phosphorus as NaH_2PO_4. The addition to such a diet of increasing percentages of aluminum sulfate up to the theoretical amount of aluminum necessary to combine with the available phosphorus limited growth to an increasing degree as the ratio of aluminum to phosphorus approached unity. In other words, insofar as phosphorus utilization of the rat is concerned, the limiting effect of this soluble salt of aluminum can be largely explained, as expected, by a simple chemical equation.

The incorporation of a powdered tablet mixture of a commercial aluminum hydroxide in the ration in such amounts as to correspond to those of the aluminum sulfate used in earlier experiments resulted in a growth rate quite materially in excess of that expected theoretically on the basis of the ratio of phosphorus to aluminum present. Under such dietary conditions approximately 30 per cent of the hydroxide salt was converted to a soluble or reactive form.

Influence of the thyroid on cyclopropane-adrenalin tachycardia. J. W. Stutzman (introduced by W. J. Meek). *Department of Physiology, University of Wisconsin Medical School, Madison.*

Cyclopropane greatly enhances the activity of adrenalin on the automatic tissues of the dog's heart. In the unanesthetized animal 0.01 mgm. adrenalin per kilogram produces only escape phenomena, while under the anesthetic it causes multifocal ventricular tachycardia in nearly every case. The present study is concerned with the influence of the thyroid on these irregularities.

Dogs were equilibrated for 30 minutes on a 30 to 32 per cent mixture of cyclopropane in oxygen. The test dose of 0.01 mgm. adrenalin per kilogram was injected at a constant rate over a 50 second period and the duration of ventricular tachycardia determined by electrocardiographic records.

Two and four weeks after thyroidectomy the above procedure was repeated. To determine the effects of hyperthyroidism on cyclopropane-adrenalin tachycardia dogs were fed 200 and 400 mgm. desiccated thyroid glands (Parke, Davis) per kilogram for 12 days and then tested by adrenalin injection under anesthesia.

Following thyroidectomy the duration of cyclopropane-adrenalin tachycardia was decreased. In the hyperthyroid state the period of tachycardia was longer than in the control.

A delayed hyper-lactacidemia following severe acute hemorrhage. Major M. Swan (introduced by O. O. Stoland). *Department of Physiology, University of Kansas, Lawrence.*

Other workers have shown an elevated lactacidemia in normal unanesthetized dogs in the first twenty-four hour period immediately following a

severe acute hemorrhage (Riegel, J. Biol. Chem. **74**: 123, 1927; Fuss, Klin. Wchnschr. **13**: 917, 1934). Our studies follow the later changes of lactic acid concentration in the blood.

Three males and three females, all in the resting and unanesthetized state, were subjected to an acute blood loss comprising twenty-eight to thirty-two per cent of the individual's total blood volume. Five cubic centimeters of peripheral venous blood were drawn at twenty-four hour intervals for determination of lactic acid by the Friedemann-Cotonio-Shaffer method. All samples were drawn at the same time each day.

Average lactic acid values in milligrams per cent on the series were: normal, 15.5; post-hemorrhage, first day, 15.9; second day, 19.8; third day, 18.7; fourth day, 12.9; fifth day, 17.4; seventh day, 11.5; ninth day, 16.4.

These results indicate a well-defined, delayed hyper-lactacidemia which occurs after twenty-four hours following severe hemorrhage. This elevated level lasts for at least a day.

The relation of morphine withdrawal symptoms in the rat to the thyroid gland. H. G. Swann. *Department of Physiology, University of Chicago, Chicago, Ill.*

Because of the similarity of certain of the symptoms in the crisis of thyrotoxicosis to those of morphine withdrawal, it was postulated that the thyroid was involved in morphine addiction and withdrawal. The theory has been found faulty in most aspects:

1. The thyroidectomized rat can be addicted to morphine if great care is exercised.

2. In the normal rat, the great weight loss (about 20 per cent of the body weight in 24 hours) following withdrawal of morphine after addiction is due partly to the constipation which addiction causes and partly to a negative water balance. The same phenomena have been observed in the thyroidectomized addicted rat.

3. In the addicted normal rat, morphine causes a considerable specific dynamic action (B. M. R.'s of about plus 20) and withdrawal of morphine is followed by a violent depression in the B. M. R. to about minus 35. The same effects can be observed in the addicted thyroidectomized rat, but they are placed around a lower base line and are less violent.

Binocular interaction and excitability cycles in cat and monkey. S. A. Talbot and W. H. Marshall. *Wilmer Ophthalmological Institute, Johns Hopkins University and Hospital, Baltimore, Md.*

We have observed excitability cycles in the geniculate-striate system in cats and rhesus monkeys under chloralose and nembutal. The anaesthetic was deep enough to measure electric response amplitude without serious interference from random effects of spontaneous central activity.

We have previously shown for monocular stimulation that the geniculate response and all cortical components show supernormality for 20 msec. followed by subnormal excitability. The supernormal phase is most marked at any anatomical level when the frequency is 1 to 5 cps., though at such frequencies both conditioning and test responses are smaller than at slow frequencies ($\frac{1}{10}$ cps.).

Experiments on binocular interaction have been done by applying conditioning shocks to one optic nerve and test shocks to the other, observing at

various levels the effect of contralateral on ipsilateral and vice-versa. The geniculate post-synaptic and the radiation responses show only the subnormal phase of interaction excitability for both cat and monkey. At the cortex the cat shows both supernormal and subnormal phases for the post-radiation components, with time relations as described for the monocular case above. As yet we have demonstrated only the subnormal phase in the cortical interaction of the monkey to electric stimulation.

Binocular interaction following photic stimulation of one eye and electric shock of the other nerve appears in the cat at the cortical level only. Responses to separate photic stimulation of the two eyes interact clearly in the monkey. At short test-flash intervals enhancement of certain cortical phases occurs, suggesting at times a supernormality imposed on subnormal excitability. The conditioning "on" response of one eye interacts with the "off" response of a long stimulation of the other, with cyclic subnormality. This suggests a common neuron for "on" and "off" response.

Binocular interaction is thus clearly present in cat and monkey, in the primary projection system below association areas, despite differences of their anatomical organization. The physiological interaction is comparable with the anatomy of reciprocal overlap of monocular and binocular paths in geniculate and cortex. The relation of such interacting recovery cycles to binocular acuity is discussed in an accompanying abstract.

The effect of indole-3 acetic acid upon the respiration of various parts of the oat seedling (Avena sativa). A. B. TAYLOR and T. W. ROBINSON (introduced by F. R. Steggerda). *Departments of Physiology and of Zoology, University of Illinois, Urbana.*

The respiration of root tips (1st 5 mm. of root), root zones (next 10 mm. of root), and coleoptile tips (1st 5 mm. of coleoptile) of 4 day old oat seedlings, in both distilled water and in distilled water to which various concentrations of indole-3 acetic acid had been added was measured with the Barcroft-Warburg respirometer.

The three parts of the seedling used different amounts of oxygen, the rate of consumption being greatest in those regions which have normally a high rate of cell division. The root tips, for example, used about 10,000 mm^3 of oxygen per gram (dry weight) per hour while the root zones and coleoptile tips consumed about 7,000 mm^3 per gram per hour.

The application of indole-3 acetic acid in high concentrations resulted in an inhibition of respiration the degree of which was dependent, at any given pH, upon the original concentration and upon the concentration of active undissociated indole-3 acetic acid present in equilibrium with its salt. Amounts of indole-3 acetic acid which caused inhibition in an acid medium had no effect in an alkaline one. The degree of inhibition seems to be correlated with the rate of respiration in the various regions studied. For a given concentration of indole-3 acetic acid, the greatest inhibition occurred in the root tips.

A study of experimental hypothalamic obesity in the rat. JAY TEPPERMAN (by invitation), J. R. BROBECK (by invitation) and C. N. H. LONG. *Department of Physiological Chemistry, Yale University School of Medicine, New Haven, Conn.*

Symmetrical, bilateral lesions were placed in the hypothalami of a litter

of four female rats by means of the Horsley-Clarke stereotaxic instrument according to the method described by Hetherington (Anat. Rec. **78:** 149, 1940). The operated animals and their litter-mate control were then observed during a period of about 10 months. Daily records were kept of food intake and body weight, and studies were made of the respiratory exchange, creatinine excretion, carbohydrate tolerance, insulin sensitivity, ketonuria, liver fat content and oestrus cycles.

Shortly after operation the operated rats began to eat approximately twice as much food as their control, and to gain weight at the rate of about 150 grams per month, as compared with 25 grams gained by the control during the same period. Another similarly operated rat which exhibited a voracious appetite after operation but was immediately pair-fed with its control has gained weight only slightly more rapidly than its normal littermate.

The basal oxygen consumption of the fat rats (whether expressed per unit of body weight, creatinine excretion, or surface area) was found to be lower than that of their control following a 25-day fast, and during the subsequent 100-day period of pair-feeding. During this time the operated rats again gained weight at a significantly more rapid rate than did the control. The fasting R.Q.'s of the fat rats were similar to those of the control, but R.Q. determinations done in the absorptive state were found to be higher in the fat rats. The daily creatinine excretion of the fat rats was found to be approximately 30 per cent higher than that of the control.

Two of the fat animals showed an apparent increase in sugar tolerance, one showed a consistently diminished tolerance, and one exhibited a progressive diminution in tolerance associated with its increase in weight. Preliminary experiments suggest that the obese animals may be less sensitive to the action of insulin than are the controls.

The obese animals showed normal oestrus cycles during fasting and paired feeding, but began to exhibit abnormal cycles when their weight reached about 550 grams.

The pancreatic secretagogue action of bile. J. Earl Thomas and J. O. Crider. *The Jefferson Medical College of Philadelphia.*

Bile in the intestine increases the flow of pancreatic juice in anesthetized cats under certain conditions (Mellanby. J. Physiol. **61:** 419) but fails to do so in unanesthetized dogs (Leuth and Ivy. J. A. M. A. **89:** 1030; Dragstedt and Woodbury. Am. J. Physiol. **107:** 584; unpublished observations by the authors). We undertook to determine whether this is a species difference or is due to experimental conditions.

The pancreatic duct was cannulated after ligating the pylorus and common bile duct in 13 cats and 12 dogs anesthetized with urethane alone, morphine and urethane (dogs only), chloralose (cats only), or ether. The effect on pancreatic secretion of injecting bile into the upper duodenum was recorded. Bile slightly increased the flow of pancreatic juice in 25 of 48 trials in dogs and in only 4 of 26 trials in cats under similar conditions. Therefore, the positive results in cats cannot be attributed to an exclusive species characteristic.

The following changes in the experimental conditions increased the percentage of positive results:

1. Section of the splanchnic nerves; 11 positive results were obtained in 12 trials in cats.

2. Mixing the bile with 10 per cent urethane; 13 positive results were obtained in 14 trials in cats.

3. Prolonged anesthesia; 17 positive results were obtained in 25 trials in dogs after 4 hours of anesthesia compared to 8 positive results in 23 earlier trials.

4. Increasing the depth of ether anesthesia almost to the point of death in both cats and dogs.

5. Circulatory and respiratory failure (followed by resuscitation) in both cats and dogs.

We conclude that bile in the intestine increases the flow of pancreatic juice only under special conditions, all of which, so far as they are known, are abnormal.

Effects of combined estrogens and androgens in the castrate rat. D. M. THOMSON and R. R. GREENE (introduced by F. T. Jung). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Ninety-six male rats were divided into 8 groups. Seven groups were castrated at 17 days of age. Six of these groups were given daily treatment for 21 days, starting 45 days after castration. Group I received 0.05 mgm. of testosterone propionate daily; group II—0.1 mgm. alpha estradiol daily; groups III–VI—0.05 mgm. testosterone propionate plus 0.0001, 0.001, 0.01 or 0.1 mgm. alpha estradiol daily; group VII—no treatment (castrate controls); group VIII—normal controls. All animals were killed on the day following the last treatment. The ventral prostates and seminal vesicles were weighed and examined histologically.

Prostates of animals receiving estradiol alone were not significantly larger than those of the castrate controls, but the seminal vesicles did show a significant weight increment. Prostates and seminal vesicles of the group treated with androgen alone were approximately the same size as those of the normal controls. Contrary to reports by other workers, the added estrogens had no synergistic or additive effect on the weights of these sexual accessories. The seminal vesicles of the groups receiving both androgens and the various doses of estrogens were not significantly different from those receiving androgens alone. The ventral prostates were smaller in all groups receiving added estrogens than in the group receiving androgen alone. They were, however, significantly smaller only in the group receiving the largest added dose of estrogen (0.1 mgm.).

Histologically the prostates and seminal vesicles of androgen treated animals resembled the normal. The added estrogens caused a depression of the androgenic effect on the prostatic epithelium. The degree of depression was roughly proportional to the estrogen dosage. Estrogen alone produced little histological effect on the prostate. In the seminal vesicles the estrogens produced no apparent modification of the androgen effect.

The relation between Brücke frequency and light-dark ratio. JAMES TOMAN (introduced by W. R. Amberson). *Physiological Laboratory, Princeton University, Princeton, N. J., and Department of Physiology, School of Medicine, University of Maryland, Baltimore.*

The author (J. Neurophysiol. **4:** 51, 1941) has reported a maximum for amplitude of flicker potentials in the human EEG at flash frequencies

approximating that of the alpha rhythm. Bartley (Psychol. Rev. **46:** 337, 1939) has reported that the flicker frequency giving maximal subjective brightness (Brücke effect) approximates that of the alpha rhythm and remains constant as light-dark ratio is varied. The present work was undertaken to examine the constancy of the Brücke frequency over a wider range of light-dark ratios. 19 experiments were performed on 5 subjects. In each experiment light-dark ratio was kept constant and several determinations of subjective brightness made for each of a number of flash frequencies up to critical fusion. Brücke frequencies (BF) were determined from the maxima of the plotted data and are tabulated below with the light phase (LP°) per cycle used.

LP°	10	15	30	45	60	90	90	90	90	90
BF	1.3$_a$	3$_a$	3.1$_a$	2.5$_a$	4.3$_a$	4.1$_a$	4.9	5.1$_a$	5.8$_b$	8.5$_a$
LP°	110	120	170	180	180	180	200	270	280	
BF	6.1$_a$	6.1$_a$	5.9$_a$	5.5	6.5$_a$	6.5	8.5$_a$	(0)$_a$	(0)$_a$	

The (a) experiments were performed on a subject previously shown to have an occasional alpha rhythm of 12 per second, the (b) experiment on a subject with a marked alpha rhythm of 10.2 per second. It would appear that the Brücke frequency does not necessarily coincide with that of the alpha rhythm. Furthermore, in spite of the scatter in the data, the Brücke frequency would seem to be a function of the light phase per flash cycle, rather than a constant. Thus the present investigation has not lent support to an interpretation of the Brücke effect in terms of a fixed cortical cycle of facilitation related to the alpha rhythm.

Permanent genital impairments in the adult rat resulting from the administration of estrogen during early life. C. DONNELL TURNER (introduced by J. William Buchanan). *Department of Zoology, Northwestern University, Evanston, Ill.* (Read by title.)

Previous workers have demonstrated that the genital impairments resulting from the administration of steroid hormones to rodents are contingent upon the amount of treatment and the age of the animal. Continued high doses of estrogen produce follicular atresia and retrogression of the corpora lutea; large doses administered for a shorter time temporarily maintain the corpora and produce a vaginal diestrus lasting for about two weeks. Varying amounts of estrogen do not luteinize the ovary of the prepuberal rat. Testosterone injections are said to result in a continuous follicular phase in the ovaries of the adult.

In the present experiments, from 100 to 200 IU of estrogen were administered daily to newborn female rats during the first ten days of life and discontinued thereafter. Some of the animals were not autopsied until ten months of age. Vaginal smears indicated that the estrous cycles were abnormal. The adult ovaries contained follicles in various stages of atresia. Corpora lutea have not been observed in the intact ovaries, but they are capable of luteinizing when transplanted to normal hosts. Some of the ovaries from adult animals of this type are capable of inducing and maintaining secretion in the sex accessories of castrate male hosts. It is believed that the androgenicity of these ovaries is correlated with the thecal hypertrophy which occurs in retrogressing vesicular follicles. Such animals

occasionally copulate during the prolonged estrous periods but do not impregnate; ovulation apparently does not occur.

Vaginal smears indicate that the treated animals remain in vaginal estrus for long periods and that the prolonged estrous periods are separated by extended intervals during which the vagina is mucified. Since corpora are absent, it is believed that the periods of mucification are attributable to diminished estrogen or to the release of an ovarian androgen which modifies the action of estrogen. The recurring periods of estrogen diminution seem to be correlated with high rates of follicular atresia.

On the basis of limited experiments, it is suggested that the estrogen produces these genital impairments by disturbing the gonad-stimulating mechanism of the hypophysis cerebri.

The respiration of the various parts of the brain during growth. DAVID B. TYLER and A. VAN HARREVELD (introduced by D. R. Drury). *California Institute of Technology, Pasadena.*

The oxygen consumption of the cerebrum, stem, cerebellum and medulla was determined in 1, 3, 5, 12, 19 and 25 day old rats and also in adult rats and mice. At birth the oxygen consumption of the various parts of the rat's brain is at a very low level and slowly increases during the next four to five weeks. During the first three weeks of life the cerebellum and the medulla take up the most oxygen per milligram of wet weight, the stem is next and the cortex is lowest. This order gradually changes and in the adult the cerebrum is highest followed by the stem, the cerebellum and the medulla being the lowest.

In the adult mouse the oxygen uptake of the various parts of the brain is greater than in those of the adult rat (per milligram of wet weight). The order of the oxygen uptake of the various regions is also different. In the mouse the cerebellum is highest.

The effect of corticotrophin on the resistance of hypophysectomized rats to low environmental temperatures. R. TYSLOWITZ (by invitation) and E. B. ASTWOOD. *Departments of Pharmacology and Medicine, Harvard Medical School, and the Medical Clinic of the Peter Bent Brigham Hospital, Boston, Mass.*

This preliminary report is based on the observation of 211 totally hypophysectomized, 31 adrenalectomized and 35 unoperated male rats, subjected to environmental temperatures of 0°, 5° and 8°C.; colonic temperature readings on treated and untreated animals were taken at hourly intervals. Under these conditions hypophysectomized animals injected for various periods with pituitary extracts rich in corticotrophin showed a striking resistance to low environmental temperatures as compared with untreated animals whose body temperature fell within a few hours to abnormally low levels. In general, it was observed that as long as the animals shivered, their temperature was well maintained, but with a disappearance of this reaction the body temperature fell and the animals showed signs of shock, coma, and death.

At 0°C. the body temperature of untreated 140 gram animals fell within 8 hours to 14°C. (their adrenals weight at autopsy 20.4 mgm.), while animals injected with a total of 100 mgm. of acetone dry powder of whole sheep pituitary during two days previous to the exposure to cold maintained a temperature of 37.2°C. (adrenal weight 24.8 mgm.). Similar

effects were obtained with three pituitary extracts in which the growth, the thyrotropic, and the gonadotropic factors had been partially removed. Four batches of pituitaries extracted by an acid acetone method yielded preparations rich in corticotrophin but free of the growth, thyrotropic and gonadotropic factors. In six series of experiments animals treated with those extracts showed a marked resistance to low environmental temperatures.

Cortin exhibited a protective effect in both hypophysectomized and adrenalectomized animals, while corticotrophin was without any effect in adrenalectomized animals indicating that the action of pituitary extracts under those conditions is mediated by the adrenal cortex.

The reaction described above is thus an index of cortical function induced by hypophyseal corticotrophin.

The effect of pantothenic acid on achromotrichia in rats. KLAUS UNNA (introduced by Hans Molitor). *Merck Institute for Therapeutic Research, Rahway, N. J.*

Achromotrichia occurs within 3 to 7 weeks in young black or piebald rats maintained on a pantothenic acid free diet consisting of vitamin free casein, dextrose, crisco, salt mixture and cod liver oil supplemented with thiamine, riboflavin, nicotinamide, pyridoxine and choline. The process of depigmentation develops in symmetrical patterns. Depilation with barium sulfide at the beginning of the experiment makes possible earlier recognition of the greying. The daily feeding of 100 micrograms of calcium pantothenate or its equivalent in dried whole liver prevents the achromotrichia. Suboptimal amounts of calcium pantothenate as well as alkali treated liver concentrates (of low pantothenic acid content) fail to prevent greying although the animals continued to grow.

The daily feeding of calcium pantothenate to rats rendered grey on this diet, causes the grey patterns to disappear gradually within 3 to 6 weeks at which time the black pigmentation is restored. Comparable effects were obtained with dried whole beef liver, whereas alkali treated liver concentrate failed to restore the pigmentation.

On similar diets but free from fat, calcium pantothenate was likewise effective in preventing or curing achromotrichia.

In the absence of pantothenic acid, the prolonged administration of hormones (pituitary, thyroid, and adrenal cortex) had no effect on achromotrichia.

The isolation of a protein from the pars neuralis of the ox pituitary with constant oxytocic, pressor and diuresis-inhibiting activity. H. B. VAN DYKE, BACON F. CHOW (by invitation), R. O. GREEP and A. ROTHEN (by invitation). *Division of Pharmacology, The Squibb Institute for Medical Research, New Brunswick, and Division of Physical Chemistry, The Rockefeller Institute for Medical Research, New York City.*

By extraction of fresh posterior lobes of ox pituitaries by a method which will be presented in detail, a protein has been isolated which has constant activity in terms of the following tests: isolated guinea pig uterus and fowl blood-pressure for oxytocic activity; blood-pressure of dog for pressor activity; inhibition of water diuresis in rats for diuresis-inhibiting activity. About 11 micrograms of nitrogen of this protein is equivalent to 1 unit of U.S.P. reference standard. In solutions of the protein (fractions low or

high in protein N as a result of electrophoretic migration), activity does not deviate from the relationship, 11 micrograms N $\eqsim$ 1 U.S.P. unit. Similarly, the activity is associated with the protein when a solution undergoes ultracentrifugation.

In melanosome-dispersing (intermedin) activity, 11 micrograms N $\eqsim$ 0.002 unit of U.S.P. reference standard.

In a solvent composed of 0.5 M acetate buffer, pH 3.94, containing 6.5 per cent NaCl, a saturated solution of the protein at 25.3° contains 0.10 mgm. dissolved N per cc. whether the solution is just saturated or whether 2 mgm. protein N per cc. are suspended in the solvent. In the Tiselius electrophoresis apparatus, solutions containing 1 per cent protein at constant ionic strength of 0.05 were found to contain one component with traces of impurity. If the latter, mixed with traces of the diffused main component, is assayed biologically, its activity has never been found to exceed that of the main component. The mobility of the protein at various pH's at 1.5° was: pH 3.4, -6.0×10^{-5}; pH 4.1, -4.2×10^{-5}; pH 5.5, $+2.9 \times 10^{-5}$, and pH 6.1, $+3.8 \times 10^{-5}$. The calculated isoelectric point is pH 4.8.

The protein behaved like a homogeneous substance in the ultracentrifuge. The following value was found for the constant of sedimentation at 6.3°, $S^{6.3} = 1.87 \times 10^{-13}$. Diffusion experiments were carried out at 0.3°. The diffusion constant was 4.4×10^{-7}. The molecular weight calculated from the diffusion constant and the sedimentation constant corrected for the difference in temperature was M = 31,000. The f/f_o value was about 1.18.

The survival of central synaptic conduction during asphyxia and anoxia. A. Van Harreveld (introduced by Gordon A. Alles). *Kerckhoff Biological Laboratories, California Institute of Technology, Pasadena.*

The period of survival of reflex action currents of the spinal cord during asphyxia is short, usually not more than 4 minutes. If, however, the spinal cord is asphyxiated for 35 minutes and then 14 days later again subjected to asphyxiation, the survival time of the reflex action currents is considerably increased. In many of the previously asphyxiated animals the reflex action currents survived circulatory arrest for 13–14 minutes. This increased survival time is not present the first few days after the initial asphyxiation, but develops in about 10 days. Histological examination of the spinal cord showed a considerable decrease of nerve cells. The total number of cells, however, was usually larger than normal due to the presence of many phagocytes. This makes rather unlikely the explanation of the increased survival time as due to a longer survival of the few remaining cells on the little oxygen reserve in the vessels and in the tissues. Furthermore, if the oxygen lack is produced by artificial respiration with nitrogen (a procedure which removes oxygen from blood and tissues), the survival times are even longer than those found when the final asphyxiation is produced by circulatory arrest. It seems therefore that the process of synaptic conduction in the central nervous system, like in the peripheral ganglia, is not very sensitive to oxygen lack.

The effect of senescence on the emptying time of the human stomach. Edward J. Van Liere and David Northup. *Department of Physiology, West Virginia University, Morgantown.*

The gastric emptying time of 12 men, whose average age was 70.7 years,

was studied. The youngest subject was 58 years and the oldest 84. Ten of the twelve were indigents residing in the county infirmary; one was a college professor; one a janitor.

A number of the subjects were reasonably robust, but several were feeble physically. They were all able, however, to do light work and could ascend a flight or two of stairs without any apparent difficulty. As far as could be determined none of them suffered from organic disease of the gastro-enteric tract.

They were given a test meal which consisted of 15 grams of Quaker Farina, cooked to a volume of 200 cc.; 50 grams of barium sulphate were added so the position of the meal could be ascertained fluoroscopically. The meal was eaten at 7:30 in the morning; no other food had been taken since the previous evening. The subjects were allowed to lounge around the laboratory and were instructed to relax mentally and physically as much as they could until the stomach was emptied. With the exception of one or two subjects, three determinations, at exactly weekly intervals, were made on each individual. The average figure was used for the norm. The time it took the meal to leave the stomach was determined to the nearest ten minutes.

It had been established previously in 49 male medical students, whose average age was about 24 years, that the same type meal under the same environmental conditions took an average of 2.11 hours to leave the stomach.

The average length of time for the meal to leave the stomach in the 12 old men was 1.94 hours. The extremes ranged from 1.33 hours to 2.75 hours. This range compared favorably with the observations made on the younger subjects.

It was concluded that the motility of the stomach of old men did not differ from that of young vigorous male adults.

Duodenal ulcer formation in the dog by intramuscular injections of a histamine beeswax mixture.[1] R. L. Varco (by invitation), C. F. Code, S. H. Walpole (by invitation) and O. H. Wangensteen. *Departments of Surgery and Physiology, University of Minnesota, Minneapolis.*

In an earlier study it was demonstrated that when histamine is placed in a beeswax mineral oil mixture its action is so prolonged, that in dogs, a single injection produces a continuous, copious secretion of highly acid gastric juice which is maintained for many hours (Proc. Soc. Exper. Biol. and Med. **44:** 475, 1940). In cats the histamine beeswax mixture likewise produces a sustained abundant flow of acid gastric juice and if administered daily results in ulceration of the stomach and duodenum (Proc. Soc. Exper. Biol. and Med. **44:** 619, 1940). The present study was undertaken to determine whether excessive gastric secretion in the dog could produce ulcers in the stomach or duodenum.

The histamine beeswax mixture was prepared by mixing finely ground histamine with one part hot beeswax and then diluting with four to five parts hot mineral oil. When homogeneous and while still molten, the mixture was drawn into a 1 cc. tuberculin syringe and allowed to solidify at room temperature.

[1] Part of the expense of this research was borne by grants from the Committee on Scientific Research of the American Medical Association.

Three normal dogs received daily intramuscular injections of this mixture in quantities which represented 30 or 40 mgm. histamine base. Following periods of four days to four weeks the animals were examined and in every instance single or multiple, perforating ulcers of one-half to one inch in diameter were found in the first portion of the duodenum.

In order to be certain that the ulcers in the duodenum were caused by the secretion of excessive amounts of acid gastric juice, two control experiments were done. In one of these, a normal dog was given daily intramuscular injections of beeswax mineral oil mixture without histamine for thirty days. At the end of this period the stomach and duodenum were normal. In the other control experiment, the beeswax mixture, containing histamine in 30 mgm. doses, was given intramuscularly each day to a completely gastrectomized dog. After thirty days of this treatment the duodenum presented a normal appearance. The duodenal ulcers produced in the normal dogs were thus due to the continuous excessive secretion of acid gastric juice evoked by the histamine in the beeswax mixture.

Increase in the protein content of the liver following a sterile subcutaneous abscess. HARRY M. VARS (by invitation), SAMUEL GOLDSCHMIDT, JULIUS SCHULTZ (by invitation) and I. S. RAVDIN. *Harrison Department of Surgical Research and the Department of Physiology, School of Medicine University of Pennsylvania, Philadelphia.*

Groups of rats were injected subcutaneously with sodium ricinoleate and fasted for 48 hours. The composition of the livers of these animals was compared with that of rats fasted and rats fed during the same period.

The liver weight of animals treated with sodium ricinoleate was greater, in many cases the total nitrogen and the total protein per liver was higher, and the total purine content was often as much as 20 to 30 per cent higher than in the fasted controls.

These differences were still greater and more consistently so after the injected animals had been anesthetized with chloroform. In many groups the total purine-nitrogen per liver of the chloroform-treated animals was close to the level found in animals fed, or starved and refed during a similar time period. Fifty-eight groups, with a total of 330 rats were used in these studies.

These dafa are a direct demonstration, by chemical analysis, of the fact that the liver is capable of utilizing, for the maintenance or growth of its own tissue, the products of tissue destruction liberated by means of a sterile abscess.

The effect of lipocaic and cholesterol administration in rabbits. CORNELIUS W. VERMEULEN (by invitation), J. GARROTT ALLEN (by invitation), DWIGHT E. CLARK (by invitation), ORMAND C. JULIAN (by invitation) and LESTER R. DRAGSTEDT. *Department of Surgery, The University of Chicago, Chicago, Ill.*

Forty male and female albino rabbits were distributed between two groups, one group receiving daily doses of cholesterol, the second group receiving cholesterol and a purified extract of lipocaic in the largest doses tolerated by the animals. The animals of the two groups were sacrificed and compared over an 18-month period. The deposition of cholesterol in the aorta of the animals was not in any way affected by the addition of

lipocaic to the diet. Other observations as to the deposition of cholesterol in other tissues were made. These results confirm the observations reported previously using cruder lipocaic preparation.

A physiological analysis of twenty-six patients with patent ductus arteriosus. ANTONIO VIOLANTE (by invitation), M. J. SHAPIRO (by invitation) and ANCEL KEYS. *Laboratory of Physiological Hygiene, University of Minnesota, and the Lymanhurst Cardiac Clinic, City of Minneapolis.*

Twenty-six patients with patent ductus arteriosus, most of whom had been under careful observation from 2 to 18 years, were repeatedly studied by physiological methods under controlled basal conditions. Ages ranged from 5 to 35, average 15.8 years; 77 per cent were females. Heart failure was not present in any of these patients, all of whom were leading relatively normal lives at the time of study. Average blood pressures were 117/59 in the arms and 140/65 in the legs. Basal pulse rate averaged 90.5. Enlargement of the pulmonary artery was slight in 30.8 per cent, moderate in 23.8 per cent and marked in 35.4 per cent. Thrill was present in 84.6 per cent. The murmur was continuous in 80.8 per cent and systolic in 19.2 per cent; sound tracings showed that the murmur is always accentuated at the second sound. The murmur was maximal in the second left interspace in 92.3 per cent and in the third interspace in 7.7 per cent. B.M.R. averaged + 4.39 per cent. Pulmonary congestion was observed in 83.8 per cent, occurring 3 times as frequently on the right side as on the left. Cardiac index (acetylene) averaged 2.27 liters per minute per square meter. Simultaneous roentgenkymography (gross stroke) and acetylene procedures indicated that the gross ventricular output averaged 25.5 per cent greater than the net circulation. The magnitude of the "leak," estimated in this way, was roughly proportional to the pulse pressure and the results were entirely similar to those obtained in patients with aortic regurgitation. Surgical closure was attempted in 3 patients with one failure, one complete success and one partial failure. The estimated "leaks" before operation in these patients were 59 per cent, 27 per cent, and 65 per cent of the gross output; the pulse pressures (arm) in these patients were 99, 59 and 84 mm. Hg, respectively.

Recovery of excitability in nerve. ERNST T. VON BRÜCKE (by invitation), MARIE EARLY (by invitation) and ALEXANDER FORBES. *Department of Physiology, Harvard Medical School, Boston, Mass.*

In frog sciatic nerve recovery of excitability after a single conditioning impulse was compared with recovery during the so-called "second" refractory state (after two conditioning impulses). Recovery was always found to be slowed down in the latter case; as the interval between the two conditioning impulses was increased, recovery in the "second" refractory period was found to become faster. This delay of recovery during the "second" refractory period may be the first detectable sign of "fatigue."

The fact that the separate refractory states after two impulses sum, just as subnormality does, and the fact that responsiveness increases no further during the second part of the refractory period are considered to favor the assumption that subnormality is a late continuation of the relative refractory state.

In studying the supernormal phase in recovery of excitability it was

found that sensory root fibers in the bullfrog do not show supernormality, whereas the peripheral sensory fibers do. Whether this represents a specific difference between the central and the peripheral processes of the spinal ganglion cell or whether an external factor is involved, has not yet been determined.

After tetanization the appearance of supernormality and of subnormality in frog sciatic nerve was delayed, the degree of delay depending upon the degree of previous activity.

The toxic factor in pernicious anemia. G. E. WAKERLIN. *Department of Physiology, College of Medicine, University of Illinois, Chicago.* (Read by title.)

In a preliminary note (Science **82**: 494, 1935), we reported that urine from eight untreated patients with pernicious anemia contained a thermolabile, comparatively toxic reticulocyte decreasing factor for the pigeon, whereas the substance was not demonstrated in the urine of six normal humans and two treated pernicious anemia patients. Since this report reexamination of the urines of three of the eight pernicious anemia patients after adequate therapy with liver extract showed an absence or marked diminution of the reticulocytopenic factor. On the other hand, the reticulocytopenic principle was found to be absent from the urines of three of five additional previously untreated pernicious anemia patients in severe relapse. Moreover, although the factor was found to be absent from the urines of one patient each with hemolytic icterus, polycythemia vera, meningococcic meningitis, and fever of unexplained origin, the urines of three patients with diffuse carcinomatosis, subacute bacterial endocarditis, and aplastic anemia, respectively, had a reticulocytopenic effect on the pigeon similar to a majority of the untreated pernicious anemia urines examined. Liver extract did not influence the reticulocytopenic action of the principle for the pigeon. Daily intramuscular injections of the reticulocytopenic substance into rabbits for three weeks produced no significant change in the blood picture. We conclude that the toxic, reticulocytopenic factor is inconstantly present in the urine of untreated pernicious anemia patients, that it is not specific for pernicious anemia but is present in other diseases, and that the factor is incidental and resultant and not pathogenetic in its relation to pernicious anemia.

Reductions in blood pressures of renal hypertensive dogs by hog renin.[1] G. E. WAKERLIN, C. A. JOHNSON (by invitation) and B. GOMBERG (by invitation). *Departments of Physiology and Physiological Chemistry, College of Medicine, University of Illinois, Chicago.*

Four renal ischemic hypertensive dogs treated for 4 months with daily intramuscular injections of hog renin, representing 1 gram of kidney equivalent per kilogram of body weight, showed striking reductions in blood pressures. In the months following therapy the pressures slowly increased until the pretreatment hypertensive range was reached. A substance (antirenin) which neutralized the acute pressor response to renin became demonstrable in the serums of these dogs. At no time during treatment or subsequently was there any evidence of untoward effects. The appe-

[1] This work was aided by a grant from the Graduate School Research Fund of the University of Illinois.

tites of the four dogs remained excellent, their weights constant and their blood urea nitrogens and urinalyses normal throughout the periods of observation. The mechanism of these reductions in blood pressure probably involves an immune (antihormone?) response to the heterologous hog renin since hypertensive control dogs similarly treated with heat inactivated hog renin and (homologous) dog renin showed no significant changes in blood pressure and failed to develop antirenin. Further control experiments now in progress will also be reported on. These include the effect of purified hog renin on renal hypertensive dogs and the effect of bilateral renal artery constriction on the blood pressure of normotensive dogs during treatment with hog renin. If the promise of these preliminary findings is substantiated by further work the effect of treatment with heterologous renin will be studied in essential hypertension in man.

A comparison of the vasoconstricting effects of renal and systemic plasmas from normotensive and hypertensive dogs.[1] G. E. WAKERLIN and M. R. SALK (by invitation). *Department of Physiology, College of Medicine, University of Illinois, Chicago.* (Read by title.)

A previous report by one of us (Proc. Soc. Exper. Biol. and Med. **41**: 51, 1939) showed that there was no significant difference between the vasoconstricting effects on surviving beef arterial rings of systemic plasmas from normotensive and hypertensive dogs. At that time we indicated that the hypothetical pressor substance of experimental renal hypertension might be present in demonstrable quantities in the venous return from the ischemic kidney but not in the systemic blood. Hence, we compared the effect of heparinized renal vein and femoral artery plasmas from four normotensive dogs on the tonus of beef arterial rings by the method of Dale and Laidlaw. The renal vein plasmas were obtained by means of London cannulae. After 10 to 14 such comparisons at weekly intervals were made for each dog the animals were subjected to bilateral constriction of the renal arteries by the Goldblatt technique. During the period of hypertension which resulted the weekly comparisons were continued on the four animals for 3, 8, 17 and 29 weeks respectively. There were no significant differences between the tone augmenting effects of the renal and systemic plasmas during the normotensive period and no evidence was obtained for any increase in the vasoconstricting effects of the renal vein or systemic plasmas during the subsequent period of hypertension. Owing to the limitations of the method employed, these negative results obviously do not rule out the presence of an as yet inconclusively demonstrated pressor substance in the renal or systemic blood of Goldblatt dogs.

Vitamins A in invertebrate eyes. GEORGE WALD.[2] *The Biological Laboratories of Harvard University, Boston, Mass., and the Woods Hole Oceanographic Institution, Woods Hole, Mass.* (Contribution No. 284.)

The retina of the squid, *Loligo pealii*, contains 1–2 μgm. of vitamin A_1, and about 3 times this quantity (measured as relative extinction in the antimony chloride reaction) of retinene$_1$ (cf. Wald. J. Gen. Physiol. **22**:

[1] This work was aided by a grant from the Graduate School Research Fund of the University of Illinois.

[2] This research was supported in part by a grant from the Josiah Macy, Jr. Foundation.

391, 1939). No trace of these or other carotenoids was found in other squid tissues.

The quantity of vitamin A remains constant in all conditions of light and darkness. Vitamin A does not appear therefore to participate directly in the visual processes.

About 15 units (relative) of retinene can be extracted with benzine in darkness. A further 7 units is released on exposure of the live squid or of the isolated retina to light. About 120 units of retinene is stored in a photostable retinal complex from which it can be extracted only with polar organic solvents like chloroform.

The squid apparently possesses therefore the simple visual cycle, visual purple $\underset{\longleftarrow}{\overset{\text{light}}{\longrightarrow}}$ retinene + (protein?). The term visual purple is used here in its generic sense. Though measurements of spectral sensitivity (Hess, Hartline) show that the squid possesses a photopigment comparable with rhodopsin, attempts to extract it have so far failed. The deep purple color of the squid retina, sometimes ascribed to visual purple, is due to a photostable, alkali-soluble, probably melanoid pigment. Other than screening the retinal cells, this probably plays no role in vision.

In the vertebrate rods retinene occupies a position symmetrical with that of vitamin A. In the squid, retinene entirely preempts the vitamin A function. It is therefore itself a vitamin A — vitamin A_3.

The eyes of green and fiddler crabs (*Carcinus maenas* and *Uca pugnax*) also contain high concentrations of vitamin A_1. No trace of retinene has been found in these tissues.

There is therefore no discontinuity between vertebrates and invertebrates in the occurrence and utilization of vitamins A in the eye. Failure heretofore to identify vitamins A in invertebrates may be ascribed to their low capacity for storing these substances.

The collection and analysis of fluid from single nephrons of the mammalian kidney. ARTHUR M. WALKER and PHYLLIS A. BOTT (by invitation), JEAN OLIVER and MURIEL C. MACDOWELL (by invitation). *Laboratory of Pharmacology, University of Pennsylvania Medical School, Philadelphia, and Department of Pathology, Long Island College of Medicine, Brooklyn, N. Y.*

In anesthetized rats, guinea pigs and opossums a technique has been developed for rendering the kidney accessible to direct puncture of its nephrons in a manner similar to that which was found serviceable in studies of the amphibian kidney. The kidney surface was protected from exposure by a layer of warm oil and illuminated by light transmitted through a lucite rod. Puncture of tubules and, in a few cases, glomeruli was accomplished by quartz pipettes and fluid was collected in amounts sufficient for ultramicroanalysis (0.1 to 1.0 c.mm.). After marking the punctured nephron by the injection of ink for subsequent identification, the kidney was fixed in formalin, macerated and the nephron isolated by microdissection in its entirety. By means of stereoscopic photographs and camera lucida drawings, accurate measurements of the various segments were then made which exactly identified the site of puncture with reference to the glomerulus.

Sixty-six successful experiments have been made. The results show that glomerular fluid is free from detectible amounts of protein, has the

same osmotic pressure as blood plasma and contains glucose (substances which reduce dinitrosalicylic acid) and exogenous creatinine in the same concentration as in plasma water. Glucose is reabsorbed by the proximal tubule, as in amphibia. The increasing concentrations of creatinine and glucose (after phlorhizin injection) in tubule fluid indicate that the proximal segment reabsorbs about 80 per cent of the fluid in glomerular filtrate; this fluid reabsorption is accomplished without any increase in osmotic pressure of the tubule contents. The tubule fluid/plasma concentration ratio of chloride reaches 1.5 early in the proximal tubule; this finding, in view of the vapor pressure observations and preliminary sodium analyses, implies the preferential reabsorption of another anion ($-HCO_3$?) by the proximal segment.

Our data on the composition of fluid in the distal tubules are too incomplete to warrant conclusions being drawn: they suggest that a change in pH but no increase in osmotic pressure occurs before the fluid reaches this segment.

The effects of local lesions of the organ of Corti on cochlear potentials. EDWARD M. WALZL and JOHN E. BORDLEY (introduced by Clinton N. Woolsey). *Otological Research Laboratory, The Johns Hopkins University, School of Medicine, Baltimore, Md.*

Minute lesions in the organ of Corti of cats have been made in a considerable number of positions along the cochlear spiral by small detachments of the spiral ligament. The exact position and character of each lesion has been ascertained by histologic sections and graphic reconstructions of the operated cochleae. The impairments in the thresholds of cochlear response were restricted to part of an octave and varied from 15 to 40 decibels depending on the size and position of the lesion. The tones affected showed close correlation with the position of the lesion, the frequency affected being inversely related to the distance from the basal end. Lesions 2 mm. from the basal end caused maximal loss for 8192 cycles, and lesions at the 14.5 mm. level gave a maximum loss for 256 cycles. The average distance along the organ of Corti for octave intervals in this range is about 2.5 mm. The tonal range explored was from 32 to 10321 cycles. Definite localization could not be demonstrated for tones below 256 cycles, and, in general, considerably less impairment was obtained for low than for high tones by lesions of identical size.

The magnitude of the impairment is related to the length of the lesion since extension of a lesion not only impaired the response specific for the area of the second lesion but also caused additional loss for the tones already affected.

These experiments demonstrate that particular regions of the organ of Corti respond optimally to specific parts of the sound spectrum, and constitute further proof that cochlear potentials are an expression of the activity of the peripheral end organ of hearing.

The mechanism of enterogastric regurgitation. JOHN WARKENTIN (introduced by A. C. Ivy). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

This investigation was undertaken to determine in more detail the mechanism concerned in enterogastric regurgitation. A total of 60 unanesthetized dogs was studied by dilute acid (0.4 per cent HCl) irrigations

of the small intestine. In the normal dog, such acid stimulations usually result in regurgitation. Three types of operated preparations were used to make the small intestine accessible: Jejunal loops under the skin; Thiry loops of the jejunum; and Thiry loops of the duodenum. All dogs had metal cannulas in the stomach, from which samples of gastric juice were withdrawn and tested for bile; such tests served as criteria of regurgitation.

We found that the introduction of acid into the intact upper jejunum will regularly induce enterogastric regurgitation after bilateral section of the splanchnics and vagi and excision of the celiac ganglion. This shows that such regurgitation can be mediated solely by the intrinsic enteric nerves. When the influence of the intrinsic nerves is avoided by applying the acid to a Thiry fistula of the jejunum, regurgitation occurred only occasionally. In dogs with Thiry loops of the duodenum, however, enterogastric regurgitation occurred in 65 per cent of tests, and in most of these dogs the vagi are essential for the response. In all these animals with Thiry loops of the duodenum the regurgitation response was significantly reduced after vagi section, which suggests that the enterogastric reflex may be concerned in this response.

The question arises as to the mechanism of regurgitation in dogs with Thiry loops of the duodenum, which still responded after bilateral vagi section. Since the regurgitation response was abolished after celiac ganglionectomy, it appears that a reflex mechanism rather than a blood-borne mechanism is involved. This suggested to us the possibility that the celiac ganglion might be a true reflex center. However, since right splanchnectomy and right lumbar sympathectomy reduced the incidence of regurgitation very markedly, it appears that the celiac ganglion is not a true reflex center for enterogastric regurgitation.

The metabolic behavior of bone marrow at low oxygen tensions. CHARLES O. WARREN (introduced by J. C. Hinsey). *Department of Anatomy, Cornell University Medical College, New York City.*[1]

This paper represents part of a program designed to investigate the physiological mechanisms involved in hematopoiesis. The metabolic behavior of bone marrow in relation to oxygen tension has been studied by two approaches; in the first, the marrows were exposed to various oxygen tensions in vitro, and in the second, the whole animal (rabbit) was exposed to lowered oxygen tension. In each case, the rates of respiration and glycolysis of slices of the marrow, suspended in serum, have been followed in the Warburg apparatus; in the first group, respiration was measured manometrically and glycolysis of the same slices was determined chemically.

In the first group of experiments, it was found that as the rate of respiration decreased with lowering of the oxygen tension, glycolysis increased in a reciprocal fashion. There was no evidence for a significant increase in glycolysis without a corresponding decrease in the rate of respiration, and in no instance did decrease in oxygen tension result in an increase in the rate of respiration. From these experiments, it is difficult to see how a lowered oxygen tension can act directly as a stimulant to marrow metabolism.

[1] The experiments reported were done during the tenure of a Lewis Cass Ledyard, Jr. Fellowship, New York Hospital 1940–41. Some of the experiments were performed at the Biological Laboratory, Cold Spring Harbor, New York.

In the second group of experiments, rabbits were placed in a tank in which the air was kept under a pressure of 410 mm., corresponding to an altitude of about 20,000 ft., thereby inducing in the course of several days, a hyperplasia of the erythroid elements in the bone marrow and a reticulocytosis and polycythemia in the circulating blood. The animals were killed at various intervals and the rates of respiration and glycolysis of the bone marrow were measured in the Warburg apparatus. The marrows were found to be characterized metabolically by a high rate of respiration in relation to the rate of glycolysis,—a type of metabolism which has been shown in earlier studies to be characteristic of red marrows in which the erythroid hyperplasia was induced by other means. These experiments indicate that the predominately oxidative type of metabolism of the immature red cells in the bone marrow is not altered when the animals are exposed to low oxygen tensions.

The influence of liver damage on the complement titer of the blood. PHILIP WASSERMAN (by invitation) and I. ARTHUR MIRSKY. *The May Institute for Medical Research, The Jewish Hospital, Cincinnati, O.*

The studies of Ecker indicate that complement is associated with the globulin fraction of blood. In view of the probability that the liver is responsible for the production of globulin, it became of interest to find the influence of liver damage on the concentration of complement in the blood.

Liver damage was produced in dogs by means of chloroform anesthesia, and the complement titer was determined before and at various intervals after the production of this damage. The complement titer was measured by determining the volume of serum necessary to produce initial hemolysis of sensitized sheep cells. The value for normal dogs was found to be 0.004 cc. serum. Within 24 hours after the induction of chloroform anesthesia, a significant decrease in the blood complement titer occurred. For the following 3 to 5 days the titer continued to drop so that up to 0.012 cc. of serum was necessary to produce initial hemolysis at the end of that time. In the succeeding 4 to 6 days, when liver regeneration is known to occur, there was a concomitant rise in the complement titer of the blood. In from 10 to 12 days after the induction of anesthesia complete restoration of the complement titer took place.

These findings suggest that the liver plays an important rôle in the production of complement.

Effects of various salts against metrazol reactions. H. WASTL. *Department of Anatomy, Hahnemann Medical College, Philadelphia, Pa.*

Intraperitoneal injection of 75 mgm./kgm. (5 per cent aqueous solution) in guinea pigs leads to very violent, alternate tonic and clonic convulsions, interspersed by periods of exhaustion in 100 per cent of the cases and death in 85 per cent. When this dose of metrazol is given in conjunction with sodium, potassium or calcium salts (all given in doses of 150 mgm./kgm., in 10 per cent solutions) injecting them intraperitoneally either prior to or in a mixture with metrazol, partial protection characterized by two features is observed: decrease in the incidence of convulsions and lowering of the mortality rates in cases in which convulsions developed. Six groups of salts used in this study fall into three groups (The mortality rate refers here to the convulsive cases). Ca-benzoate and Ca-salicylate

lower the incidence of convulsions by roughly 40 per cent and have almost no influence on the mortality rate. $CaCl_2$ and Ca-levulinate diminish the incidence of convulsions by about 30 per cent and the mortality rate by about 12 per cent. Ca-gluconate and Ca-lactate decrease the incidence of convulsions by 20 per cent and the mortality rate by 35 per cent. In order to analyze further the anion and cation effects on the main site of action of the drug (i.e., central nervous system) the Na, K, and Ca forms of these salts were compared. The following examples illustrate the invariable trend, observed in over 1500 experiments. Injected separately immediately before metrazol: with Na-levulinate 37.5 per cent not very violent convulsions, mortality 25 per cent (whole group), 66.6 per cent (convulsive cases); with K-levulinate 43.8 per cent not very violent convulsions, mortality 33.3 per cent (whole group), 76.2 per cent (convulsive cases); with Ca-levulinate 70.8 per cent convulsions, fairly violent, mortality 56.2 per cent (whole group), 79.4 per cent (convulsive cases); with Na-gluconate 48.4 per cent not very violent convulsions, mortality 28.5 per cent (whole group), 61.3 per cent (convulsive cases); with K-gluconate 52.1 per cent medium strong convulsions, mortality 29.2 per cent (whole group), 56.0 per cent (convulsive cases); with Ca-gluconate 75.0 per cent fairly violent convulsions, mortality 33.3 per cent (whole group), 44.4 per cent (convulsive cases). Similar results—namely that with separate injections the protective effects of the sodium form of the salts are best, of the calcium form least, the potassium form occupying an intermediate position—were observed in extensive studies with local anesthetics, used in convulsant doses.

The properties of protein fibers produced reversibly from soluble protein molecules. DAVID F. WAUGH (introduced by Francis O. Schmitt). *Washington University, St. Louis, Mo.*

A 2 per cent solution of insulin hydrochloride (Lilly) in distilled water heated at 100°C. for 30 minutes forms a thixotropic gel which, after an initial disturbance, shows strong static birefringence (I. Langmuir and D. F. Waugh. J. Am. Chem. Soc. **62,** 1940). Dilution of the 2 per cent gel to 0.7 per cent causes flow birefringence to replace the static birefringence. The flow birefringence, positive with respect to the direction of flow (and other evidence to be reported) shows that anisodiametric rodlets or fibers are involved. The low rates of shear necessary to produce birefringence indicate that the asymmetry factor is high.

During drying, the optical properties of a film of the birefringent gel change in a manner which indicates that form birefringence is involved, the possible component of eigen birefringence being too small to measure. The bearing of these facts on the possible structure of the insulin molecule will be discussed.

Considerable reversibility is indicated by disappearance of birefringence and decrease in viscosity after a multiple freezing-thawing treatment since a subsequent heat treatment causes the birefringent gel to reform.

Jensen (*Insulin, its chemistry and physiology.* London, Oxford Press, 1938) states that the heat precipitate of insulin, when dissolved in dilute alkali, exhibits practically the same activity as the original material. This fact, together with the above experiments and force-area data, indicate that heat treatment does not radically alter the globular nature of the insulin molecules.

Results similar to those for insulin have been obtained for at least one other protein: crystalline egg albumin.

The formation of protein fibers from soluble protein molecules is of fundamental biological interest. The reversible formation of the spindle and asters in living cells and the irreversible formation of fibrin from fibrinogen, all of which show birefringence positive with respect to the fiber axis, are pertinent examples. It is suggested that there is a fundamental similarity between the forces which maintain the directional molecular aggregation in the insulin gel and the forces which maintain many reversible and non-reversible directional aggregations in vivo, although the processes by which the aggregations are brought about may be expected to be different.

The mechanism of ventricular fibrillation after digitalis.[1] RÉNE WÉGRIA, J. H. GEYER and B. S. BROWN (introduced by C. J. Wiggers). *Department of Physiology, Western Reserve University, Medical School, Cleveland, O.*

The question whether or not digitalis induces fibrillation by the same mechanisms as electric currents and ischemia was studied.

The fibrillation threshold was determined by measuring the strength of a D. C. shock (0.01–0.02 sec.), which, applied during the vulnerable period, is just sufficient to induce fibrillation.

It was found that digitalis and ouabain, in doses sufficient to elicit clear signs of action, do not alter significantly such a fibrillation threshold.

Further studies showed that the type of fibrillation induced differs significantly from that caused by electric currents or ischemia. After development of nodal and premature ventricular beats and occasional paroxysms of ventricular tachycardia, the E.C.G. deflections show pronounced widening and these are accompanied by little elevation of ventricular pressure. Finally, these become irregular and lead to a true state of coarse fibrillation.

The conclusion is reached that the onset and development of such fibrillation is due to favorable development of localized blocks and changes in cardiac conductivity and does not require the advent of an effective stimulus during the vulnerable period.

Studies on the aqueous humour. C. B. WELD, H. DAVSON (by invitation) and W. H. FEINDEL (by invitation). *Department of Physiology, Dalhousie University, Halifax, Nova Scotia.*

The distribution of sodium and chloride between the aqueous humour and blood serum of dogs has been investigated. The observation (T. H. Hodgson, J. Physiol., **94:** 118, 1938) that the chloride is concentrated in the aqueous humour to a greater extent than is demanded by the Gibbs-Donnan equilibrium has been confirmed. It is not due to the use of cocaine, inhalation or injection anaesthesia, nor is it due to the presence of the lens, or to evaporation from the cornea. After paracentesis it is found that there is at first no excess of chloride in the newly formed aqueous humour; within a period of about six hours, however, the excess becomes as great as it was before paracentesis. On the other hand the distribution of sodium between the aqueous humour and blood is in conformity with

[1] Aided by a grant from the John and Mary R. Markle Foundation.

the requirements of the equilibrium, so that if the excess of the chloride in the aqueous humour is to have any osmotic significance it must be balanced by some cation other than sodium. An important means of investigating the nature of the membrane separating the aqueous humour from the blood is given by a study of its permeability or otherwise, to inulin, and experiments will be described on the effects of injection of inulin into cats and dogs on the reducing value of the aqueous humour.

The effect of enterectomy on gastric secretion.[1] J. A. WELLS (by invitation) and JOHN S. GRAY. *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

A continuous hypersecretion of acid gastric juice (114 cc. containing 312 mgm. of HCl per 6 hrs.) has been observed following the removal of the small intestine of the dog. The purpose of the present study was to investigate the possibility that this hypersecretion was due to the surgical elimination of the enterogastrone mechanism.

The entire small intestine (pylorus to the ileocolic sphincter) was removed from 13 female dogs. In addition, pancreatic duct ligation and external drainage of the gastric and biliary secretions was established. Eleven female control animals were operated in the same way, except that the small intestine was not removed from the body.

The gastric fistula bags were emptied, and the animals were catheterized every 3 hours throughout the 4 to 7 day observation periods. The animals were given subcutaneous administrations of modified Locke's solution, and the biliary fistula bags were emptied every 6 hours. The gastric samples were centrifuged and the supernatant fluid was titrated against standard base, using Töpfer's as the indicator.

The results of this experiment indicate that a marked hypersecretion occurs in both the enterectomized (114 cc. containing 312 mgm. HCl per 6 hr. period) and the control (133 cc. containing 358 mgm. HCl per 6 hr. period) animals. This hypersecretion has continued in all dogs for at least 4 days, and in some cases as long as 2 weeks. The hypersecretion parallels the fluid intake. Atropine administration decreased but did not abolish the hypersecretion of acid, indicating that the pathological liberation of histamine may contribute to the hypersecretion.

The observed hypersecretion appears to be related to the stimulatory effects of surgical intervention and postoperative care rather than to the surgical elimination of the enterogastrone mechanism.

Is pectin metabolized? S. C. WERCH (introduced by J. M. Beazell). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

Our previous work has shown that the breakdown of pectin occurs in the colon and that it is probably brought about by bacterial action.

Pectin is a hemicellulose, and on acid hydrolysis yields galacturonic and acetic acids, galactose and methyl alcohol. Since similar decomposition may take place in the colon, we have approached the question, "Is pectin metabolized?" by analyzing urine and feces, from humans fed with pectin, for galacturonic and volatile fatty acids, and reducing substances as a key to the fate of the above possible products.

[1] Aided in part by a grant from the Committee on Endocrinology of the National Research Council.

The pectin used was the same pure citrus pectin used in our previous work, obtained from the Research Laboratory of the California Fruit Growers' Exchange. This product is essentially free of such impurities as pentoses, pentosans, color and other materials frequently found in other pectins.

Normal individuals were placed during a control absorption period on a diet low in crude fiber. The test period included 30 grams of pectin per day in gelatin capsules. Each period was four days in length. Aliquots of feces and urine for analysis were obtained from material secured during the third and fourth days of each period.

Fresh urine was used for the estimation of galacturonic acid and free reducing substances, and the procedure was carried out the day the specimen was secured. Determinations for total hydrolyzable reducing substances and volatile acids, on urine and feces, were made on material which had been hydrolyzed by heating in a boiling water bath for 6 hours with 2 N sulphuric acid. Galacturonic acid was estimated by the Kapp method, reducing substances by the Hagedorn-Jensen and Somogyi, Shafer, Hartman methods; and volatile acids by the Friedemann method.

Results obtained by the above procedures indicate that no significant difference exists in the excretion of galacturonic and volatile acids, and reducing substances during the control and test periods. Thus, the products of pectin decomposition cannot be detected in the urine and feces.

Initiation of shock through loss of blood or plasma.[1] JACOB M. WERLE and RICHARD S. COSBY (introduced by Carl J. Wiggers). *Department of Physiology, Western Reserve University Medical School, Cleveland, O.*

Can loss of plasma or whole blood *per se* lead to irreversible shock? This problem was studied by recording carotid pulses optically in dogs anesthetized with morphine and sodium barbital. Mean pressure was lowered to 40 to 70 mm. Hg. for two hours by one to eight hemorrhages or by acute plasmapheresis.

Some dogs developed irreversible shock as indicated by failure not only of the arterial pressures but also of the contours of arterial pressure pulses to return to normal and to remain there for several hours following reinjection of all the blood or plasma withdrawn. Other dogs recovered completely though subjected to the same procedures.

During the low blood pressure period after hemorrhage or plasmapheresis in all dogs and also after transfusion in those dogs in irreversible shock, the arterial pressure curves were small in amplitude with a prominent decline of systolic pressure; they showed a slow rise to a midsystolic peak followed by a systolic collapse. Sometimes the onset of ejection caused a huge primary oscillation. The incisura was low and the diastolic limb almost horizontal. The difference between integrated systolic and diastolic mean pressures remained normal during shock.

The dogs which did not recover completely upon restoring all the fluid withdrawn, did however experience a half-hour improvement therefrom. The reasons for the poor response are obscure. It is not due to visceral engorgement nor to withdrawal of the reinjected fluid by the spleen—as determined by autopsy and by means of splenic area measurements.

[1] Aided by a grant from the Commonwealth Fund.

The length of the low pressure period undoubtedly plays an important rôle in preventing recovery.

Simultaneous strong faradic stimulation of both central vagi, in some dogs with low pressures, raised the pressure to 220 mm. Hg, although the pulse contours remained abnormal. However, those dogs which did not respond to vagal stimulation still developed high pressures with greatly improved pulse contours after adrenalin injection.

pH changes in the blood following sulfapyridine and sulfathiazole administration.[1] GRACE E. WERTENBERGER (introduced by E. M. Greisheimer). *Department of Physiology, Woman's Medical College of Pennsylvania, Philadelphia.*

The change in blood pH after single intraperitoneal injections of varying amounts of the sodium salts of sulfapyridine and sulfathiazole was studied in a series of young albino male rats of a standard strain and adult Wistar female rats. The diet was constant and the animals average about 200 grams in weight. The pH was determined electrometrically with the Beckman potentiometer using the Behrmann-Fay blood chamber for anerobic determinations with the glass electrode and collecting the blood by cardiac puncture. Readings were made just before and three hours after administration of the drug, the latter being given in doses of 1 cc./100 grams of body weight.

A solution of 10 per cent sodium sulfapyridine caused an average rise of 0.134 point in the blood pH from an average control pH of 7.52 to an average pH of 7.66 after injection, the range after injection being 7.61 7.74. A 10 per cent solution of sodium sulfathiazole also swings the pH toward the alkaline side from an average control pH of 7.53 to an average pH of 7.60 after injection.

A 7.5 per cent solution of sodium sulfathiazole gives similar results showing a rise from the average control pH of 7.53 to an average pH of 7.63 after injection. Preliminary experiments on 7.5 per cent sodium sulfapyridine also show a swing toward the alkaline side.

Uninjected animals run as controls with each of these groups showed no change or a slight fall (0.02 point) after the 3 hour interval. This slight fall might be expected since all animals were fasted for seventeen hours before experimentation.

These data support the view that the high alkalinity of these compounds tends to exhaust the buffer capacity of the body and produce an alkalosis when given in large doses.

Estrogen excretion in hormone-induced menstrual cycles in an ovariectomized woman. N. T. WERTHESSEN (introduced by G. Pincus). *Endocrine Laboratory of the Boston Dispensary and the Joseph H. Pratt Diagnostic Hospital, Boston, Mass.*

Menstrual cycles were induced in an ovariectomized girl by the use of estradiol benzoate. The estrogen excretion was studied during the period of injection and menstruation.

It was found that approximately 10 per cent of the injected material was excreted in the urine as active estrogen when the first 48-hour period after

[1] Supported by a grant from the Committee on Therapeutic Research, Council on Pharmacy and Chemistry, American Medical Association.

injection was examined. After prolonged injection periods the amount of unabsorbed estrogen was such that only a small decrease in quantity of output was noticed when collection was delayed 24 to 48 hours after injection.

During menstruation and pregnenilolone therapy the estrogen titre of the urine fell below the values obtained before hormone administration. Estradiol, estrone and estriol fractions were found active both before and after injections had begun. The activity of the estriol fraction was lowest during menses.

Thirty milligrams of estradiol in 2 fifteen-milligram pellets were implanted. One cycle was induced by the administration of 4 mgm. of estradiol 3 weeks after insertion of the pellets. A six-week cycle followed without the administration of more hormone. Three 24-hour urine specimens collected at weekly intervals during the 3 weeks following implantation were analysed. Nineteen gamma of estrone equivalent was the highest urinary value obtained.

Menses were always accompanied by dysmenorrhea unless at least 30 mgm. per day of pregnenilolone had been taken orally for 10 days.

Hypophysis and renal function. H. L. White, Peter Heinbecker and Doris Rolf (by invitation). *Departments of Physiology and Surgery, Washington University School of Medicine, St. Louis, Mo.*

The effects 1, of complete hypophysectomy (includes destruction of median eminence); 2, of "simple" hypophysectomy, and 3, of destruction of the neurohypophysis only (including median eminence) on diodrast and inulin plasma clearances and renal extractions, on renal plasma flow, on tubular maximum rate of diodrast excretion, and on blood volume, have been followed for various periods in a series of dogs. Within a few days after either simple or complete hypophysectomy there is a marked fall in clearances, in renal plasma flow and in diodrast tubular maximum. Recovery of these functions has not yet been seen. There may be some fall after destruction of neurohypophysis only, but it is hardly beyond the normal limits of variation; the major part of the effect after hypophysectomy is due to loss of the anterior lobe. Renal extraction of diodrast is not significantly lowered; this finding along with lowered renal plasma flow and diodrast Tm may mean that the increased time of passage of blood through the kidney permits normal extraction in spite of an impairment in the power of the tubules to excrete diodrast. The lowered renal plasma flow is not due to closure of renal vessels, as is indicated by the finding that all renal vessels are injected by ink.

A working model of the crossing caval blood streams in the fetal heart. William H. Whitehead (introduced by W. F. Windle). *Department of Anatomy, Northwestern University Medical School, Chicago, Ill.*

(Demonstration.)

Physiological experiments showing that the blood streams from superior and inferior venae cavae cross in the fetal right atrium with little or no admixture have been reported (Anat. Rec. **77:** 417). A "neoprene" latex reconstruction of the right atrium and its apertures was prepared from the heart of a full term cat fetus. A stream of blue fluid admitted through the superior vena cava of this model emerges from the atrioventric-

ular orifice, while a stream of red fluid admitted simultaneously through the inferior vena cava emerges from the foramen ovale. There is little admixture of the colored fluids when the pressure in the inferior caval stream is about 20 mm. Hg greater than that in the superior. Although the model does not duplicate all conditions in the living heart, it does demonstrate that one should not assume that two streams of blood entering a common chamber necessarily mix.

Further experiments on the origin of urogastrone.[1] E. WIECZOROWSKI (by invitation), JOHN S. GRAY, C. U. CULMER (by invitation) and J. A. WELLS (by invitation). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

In a previous report of a preliminary nature, it was stated that urogastrone behaved as though it were excreted enterogastrone in that it practically disappeared from the urine of dogs subjected to a complete enterectomy. These experiments have been repeated and extended with different results as herein reported.

Seven series of experiments, each of which included at least 3 dogs, have been performed. In five of the series the output of urogastrone (on a kilo-hour basis) was determined both before and after an operation consisting of removal of the entire small intestine, external drainage of gastric and biliary secretion, and obstruction of pancreatic secretion. In two other series the output was determined before and after a control operation in which gastric, biliary and pancreatic secretion were diverted from the small intestine.

In one series of enterectomized animals the output of urogastrone was nearly abolished, in another it was reduced and in the remaining three it remained essentially unchanged. In both of the control series the output was increased.

These results reveal that removal of the small intestine does not uniformly reduce the recovery from the urine of substances which inhibit gastric secretion, although the exclusion of digestive secretions from the intestine appears to augment it. Hence, if there is only one substance in the urine extracts which affects gastric secretion, it does not originate from the small intestine alone.

The circulatory response of the unanesthetized dog to adrenalin. HAROLD C. WIGGERS, A. M. DUSCHATKO (by invitation) and R. C. KORY (by invitation). *College of Physicians and Surgeons, Columbia University, New York City.*

Experiments were conducted on six trained, normal unanesthetized dogs. Adrenalin was administered intravenously in amounts varying from 0.3 to 3.0 gamma per kilo. Femoral arterial pulse pressures and rates were measured with a modified Gregg optical manometer system.

Systolic pressures rose considerably more than diastolic with doses of 0.83 to 2.5 gamma per kilo. With smaller doses, where vagal slowing was less, diastolic elevation kept better pace with the systolic rise. Maximal diastolic elevation was attained with 2.5 gamma. Systolic pressures, on the other hand, were still increasing, though more gradually, with 3.0

[1] Aided in part by a grant from the Committee on Endocrinology of the National Research Council.

gamma/kilo. An initial tachycardia, present with any dose, was always reversed to bradycardia before sustained maximal pressure elevations were attained. The reversal occurred at approximately the same degree of tachycardia with all doses. The rate at which maximal slowing occurred varied directly with the dose, the degree varying logarithmically with the dose through the 2.5 gamma range. Arrythmias interfered with determinations when stronger doses were employed. With any dose the degree of cardiac slowing appeared to vary directly with the initial heart rate but the pulse pressure elevations exhibited no definite correlation with initial pressure levels. Maximal systolic elevations were always attained sooner and were less enduring than maximal diastolic rises. The peak of cardiac retardation was reached even later in many instances. The ascending and descending slopes of the systolic responses were steeper than those of diastolic and heart rate responses.

Several investigators have shown that in man there is no significant cardiac slowing or elevation of diastolic pressure in response to intravenous injections of adrenalin in amounts varying from 0.1 to 0.7 gamma/kilo. Myer and Spiegelhoff (Arch. f. exp. Path u. Pharmakol. **190:** 256, 1938) suggest that the reacting mechanisms differ in man and dog. The large and consistent rise in diastolic pressure in the dog indicates that the peripheral vasoconstrictor action of adrenalin must still be looked upon as contributing materially to the pulse pattern along with an increase in cardiac output and a likely reduction of aortic capacity.

Can sediment be "washed out" of the gall bladder? H. S. WIGODSKY and B. P. PHIBBS (introduced by A. C. Ivy). *Department of Physiology and Pharmacology, Northwestern University Medical School, Chicago, Ill.*

In order to obtain some indication of the flow of bile through the gall bladder, and to answer the question referred to in the title, an inert insoluble substance was placed in the gall bladder and the time necessary to wash this substance out of the gallbladder was observed. Sand was found to empty well from the gall bladder and could be followed radiologically.

Four groups of dogs, each consisting of 15 animals, were used in this experiment. Ten grams of washed, screened lake sand were placed in the gall bladder of each dog at operation. The feedings were begun as soon as possible. Feedings and treatment were given four times daily. The following feedings were given and results obtained: *a.* Canned dog food: 17.5 days to empty. *b.* Canned dog food + 2 ketochol tablets: 16.8 days to empty. *c.* One hundred and fifty cubic centimeters of equal parts of whole eggs and cream: 9.6 days to empty. *d.* One hundred and fifty cubic centimeters of equal parts of whole eggs and cream, + 2 ketochol tablets: 8.1 days to empty.

The difference in the emptying time of the sand in groups *a* and *b* on the one hand, and *b* and *c* on the other, is statistically significant. A choleresis *per se* does not "flush" the gall bladder. Repeated evacuation of the gall bladder does "flush" the gall bladder.

The origin of "off-responses" in the optic pathway. A. WILSKA (by invitation) and H. K. HARTLINE. *Department of Physiology and Biophysics, Cornell University Medical College, New York City.*

A characteristic of the activity of the vertebrate optic nerve is the strong discharge of impulses occurring in response to cessation of illumination. Analysis shows that many of the individual fibers are inactive during illumination of the retina, and respond only when the light is turned off. It is not known whether these "off-responses" originate in special receptor cells or whether they have their origin in the ganglionic layers of the retina. In the lateral eye of *Limulus* where the optic nerve fibers are axons of the primary receptor cells, the only response is a continuous discharge of impulses during illumination; "off-responses" are never observed. The ganglionic elements in the Limulus optic pathway are found in the optic lobes of the central ganglion. We have explored this region by means of micro-electrodes to determine whether types of responses similar to those obtained from vertebrate retinal ganglion cells are to be found.

In the anterior portion of the optic lobe containing the terminations of the optic nerve fibers activity is recorded during illumination of the eye, similar in all respects to that recorded from the optic nerve itself by ordinary methods. Posterior to this region we have succeeded in recording discharges of impulses in response to cessation of illumination. These "off-responses" are similar to those observed in the vertebrate retina. There is no activity of these neurons during illumination of the eye; the number of impulses and the frequency of the discharge in response to turning off the light depends upon the intensity and duration of the preceding illumination. The discharge is inhibited by re-illumination. Short periods of electrical stimulation of the optic nerve (10 to 100 shocks per sec.) cause discharges of impulses in these same elements upon cessation of the stimulation. This excludes the possibility of the peripheral origin of the "off-responses" in *Limulus*.

These observations show that certain nerve cells may discharge impulses only in response to the cessation of preganglionic stimulation.

Reactions of the anesthetized dog's chemoceptively-deafferented respiratory mechanism to hypoxemia. CLAUDE V. WINDER and HARRIET O. WINDER (by invitation). *Department of Physiology, University of Michigan, Ann Arbor.*

One hundred and sixty-eight administrations of N_2 and 6 per cent or 10 per cent O_2 in N_2 to 16 dogs (morphine, and urethane or chloralose) with carotid and vago-aortic nerves cut have been recorded. Typical reactions, short of failure, *during* the administration, were 1, progressive decrease in amplitude or amplitude and rate of breathing; 2, decrease in amplitude with acceleration of various degrees; 3, decreased amplitude with more or less acceleration and partial or complete subsequent recovery of amplitude, and 4, acceleration with very little change or with increase in amplitude. Under ordinary anesthesia and at body temperatures of 34.2–40.0°C., 83 per cent of responses were type 2 or 3. With one exception the small number of type 1 responses could be converted to 2 or 3 by prolonging or intensifying the hypoxemia. There were few of type 4. In this large group of type 2 and 3 responses, the acceleration and average reduction in amplitude resulted in small net increases in minute-volume in about a third, indefinite effects in about a fourth, and decreases in the remainder of cases, *during* the administration.

In contrast with this, during body temperatures of 40.0–42.8°C. minute

volume increased during all administrations except 4 in two dogs which had received extra anesthetic. Stimulation was sometimes striking. The majority of responses were type 4, the remainder type 2. This effect of temperature was reversible.

Irrespective of net minute-volume changes, the first few breaths of room air following an administration commonly resulted in abrupt, temporary depression which could be shown to be independent of incipient failure by varying the duration of administration. It appears to represent quick reduction of an excitatory component while depression components are still operating.

More or less prolonged after-stimulation was typical. It may represent a combination of delayed recovery from depression components and arrival of blood-borne excitants. The over-all hypoxic response thus shows an even stronger set of excitatory components than is apparent during the administration only.

The data suggest that even in anesthetized animals central excitation may operate with chemoreflex in combating central depression.

Rôle of carbon dioxide in resuscitation at birth after asphyxia and after nembutal anesthesia. W. F. WINDLE and R. F. BECKER (by invitation). *Department of Anatomy, Northwestern University Medical School, Chicago, Ill.*

Pregnant cats and guinea pigs near term were subjected to anemic decerebration or local anesthesia to facilitate observations in unanesthetized fetuses. After delivering one fetus for control, litter mates were asphyxiated by clamping the umbilical cord or uterine vessels. In other experiments, after delivering one fetus, nembutal was administered to the mother in dosage smaller than required for adult anesthesia; no further attempt was made to induce asphyxia in these fetuses.

Intrauterine respiratory movements appeared only when placental exchange was impaired. Fetuses delivered without anesthesia and without asphyxia began to breathe as soon as the face reached the air. Afferent stimulation aided in initiating respiration.

Asphyxia induced marked intrauterine activities including fetal respiratory movements. Rate and amplitude of fetal respirations indicated degree of fetal asphyxia. Cat fetuses were more resistant to asphyxia than guinea pigs. Asphyxia was carried as far as compatible with survival after delivery (some showed symptoms of central nervous system damage subsequently). Newborn animals were resuscitated in warm chambers containing air-oxygen atmospheres. Admission of about 5 to 10 per cent carbon dioxide usually proved beneficial, often increasing depth and rate of respirations.

Nembutal, insufficient to anesthetize the mother completely, produced marked depression of the fetal nervous system. At delivery the fetus failed to breathe immediately and resuscitation was accomplished with difficulty. Air-oxygen mixtures facilitated resuscitation. Some guinea pigs responded to carbon dioxide but no newborn kitten was benefited by this gas. In several experiments litter mates delivered simultaneously after nembutal administration were placed in air-oxygen and air-oxygen-carbon dioxide mixtures respectively; those in the latter atmosphere died while those receiving no carbon dioxide recovered.

Nembutal is transmitted across the placenta of the cat and guinea pig. It appears to raise the respiratory center threshold to such a high level (especially in the cat) that carbon dioxide and afferent nerve impulses have difficulty in reaching the center to initiate breathing at birth. Resuscitation with carbon dioxide is contraindicated when nembutal has been administered during labor.

Effect of whole bile and various bile constituents on gastric motility of the dog. JAMES M. WINFIELD and JERZY KAULBERSZ (introduced by Charles G. Johnston). *Department of Surgery, Wayne University College of Medicine, Detroit, Mich.*

While studying the effects of feeding dried whole bile to patients with a variety of lesions, it was noted that the symptom of anorexia was often relieved. In the following study it was found that when whole bile from various sources, or bile salts dissolved in water were placed in a fasting dog's stomach during the quiescent phase, gastric hunger contractions were invariably produced. On the contrary, when these substances are placed in a fasting dog's stomach during the contraction phase, there occurred a relatively short but definite inhibition of contractions in approximately three-fourths of the experiments. In addition, it was found that choline in a concentration found in dried bile did not call forth contractions, and produced inhibitions of contractions approximately as did the bile. Several magnesium salts produced contractions in approximately 50 per cent of the experiments and caused inhibition of contractions in about three-fourths of the experiments. Potassium chloride, on the other hand, produced definite contractions during the quiescent phase, and diminished the tone during the contraction phase.

The motility of the dog's stomach was then recorded during the feeding of canned dog food, beef heart, gelatin, bacon, olive oil, and sugar. Shortly after the feeding of fat, either as bacon or olive oil, hunger contractions ceased. In only 2 of 24 experiments did the feeding of whole bile after the intake of fat call forth contractions after a resting phase produced by fat. The introduction of gelatin and sugar caused a cessation of gastric hunger contractions, and the feeding of mixed dog food produced feeble digestive contractions. The subsequent introduction of bile did not affect the above results.

The feeding of beef heart, produced digestive gastric contractions similar to type I hunger contractions. When whole bile was introduced, with the beef heart, the contractions were diminished, but occasionally a rise of tone was noted after the feeding of whole bile.

Toxicity of potassium in adrenalectomized dogs.[1] A. W. WINKLER, H. E. HOFF and P. K. SMITH (by invitation). *Department of Internal Medicine and Laboratories of Physiology and Pharmacology, Yale University School of Medicine, New Haven, Conn.*

Potassium salts were injected intravenously into adrenalectomized dogs which had been maintained on salt without cortin. The range of concentration of potassium in serum at the moment of death was identical with

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that required to kill normal animals. Electrocardiographic events prior to death were of the same character and occurred in the same sequence in adrenalectomized and in control animals. There was therefore no evidence of increased or altered sensitivity of the heart to potassium in the adrenalectomized animal. Nevertheless much smaller amounts of potassium sufficed to kill the adrenalectomized animals. This may sometimes have been due in part to the presence of an initally elevated concentration of potassium in the serum. Diminished excretion of potassium by the kidneys was regularly a contributory cause. However, the most important cause from a quantitative standpoint was the greatly diminished apparent volume of distribution of the injected potassium, so that the adrenalectomized animals required less than one third as much potassium as did normal animals in order to produce an equivalent increase of concentration in serum.

The relationship between urinary total nitrogen and the polyuria of experimental diabetes insipidus. CHARLES A. WINTER and W. R. INGRAM. *Departments of Physiology and Anatomy, State University of Iowa, Iowa City.*

Normal cats fed a stock diet composed of raw ground lean beef and milk, and containing 3.2 grams of nitrogen in a daily portion, excrete urine with a total nitrogen concentration of about 25 mgm. per cubic centimeter on the average. Cats in the permanent phase of diabetes insipidus following interruption of the supraoptico-hypophyseal tracts concentrate the urinary nitrogen only to the extent of 4 to 10 mgm. per cubic centimeter, depending upon the degree of polyuria. Diets have been devised containing various amounts of nitrogen, from about a third the amount in the stock diet, to about 50 per cent more than the stock diet, but the caloric value and NaCl content kept appproximately constant. The normal cat responds to the high nitrogen diet by greatly increasing the nitrogen concentration in the urine, sometimes to as high as 50 mgm. per cubic centimeter or more, and there may or may not be a slight increase in urine volume. The d.i. cats, on the other hand, show only slight or no increased urinary nitrogen concentration, but show a marked increase in urine volume, so long as free access is allowed to drinking water. On a low nitrogen diet, there is some reduction in urine volume of the normal cats, but the reduction in the polyuria of the d.i. cats is most striking and dramatic, and the effect is independent of the caloric or NaCl intake, at least within fairly wide limits. At all levels of urinary nitrogen excretion, there is a close parallelism between total urinary nitrogen and urine volume in the d.i. cats. A d.i. cat completely deprived of water and fed the stock diet calls upon his reserve of body water to help excrete the nitrogen, and hence runs a strongly negative water balance and eventually shows signs of extreme dehydration. Under these circumstances, the d.i. cat is able to effect a concentration of urinary nitrogen greater than that which he would ordinarily exhibit, but at best it never approaches the level found in the normal cat.

Conditions influencing the course of steroid hormone anesthesia. HELEN WINTER (by invitation) and HANS SELYE. *Department of Anatomy, McGill University, Montreal, Canada.*

Experiments in the rat indicate that the anesthetic effect of intraperitoneally administered progesterone is considerably more pronounced in female rats than in males of the same age and weight. The probable reason of this sex difference in the responsiveness will be discussed.

Subcutaneous administration of large amounts of glucose tends to counteract the anesthetic effect of intraperitoneally administered progesterone while insulin exerts an opposite effect.

The effect of liver damage by carbon tetrachloride on fatty acid utilization of rats receiving a fat-free diet. IRWIN C. WINTER (introduced by Lathan A. Crandall, Jr.). *Department of Pharmacology, University of Oklahoma School of Medicine, Oklahoma City.*

The fatty acid "loss" of two groups of male albino rats over a two week period was determined by the paired feeding method previously described (J. Biol. Chem. **135:** 123, 1940). Twelve pairs of litter mates made up each group, one litter mate of each pair serving as a normal control, the other receiving 0.05 cc. of carbon tetrachloride per 100 grams of body weight every other day. Group I received a fat-free diet (J. Nutrition **17:** 115, 1939) plus 10 per cent ethyl stearate; group II the fat-free diet alone. Adequate vitamin supplements including linoleic acid were incorporated into the diets.

The average fatty acid loss of the normal animals of group I was 4.21 $\pm$ 0.11 grams, and of the carbon tetrachloride rats, 2.98 $\pm$ 0.10. The normal animals lost an average of 14 grams body weight, the treated rats 8 grams. On the fat-free diet the fatty acid loss was much decreased as compared to Group I, and was not significantly decreased further by the administration of carbon tetrachloride (normal = 0.74 $\pm$ 0.09 grams; treated = 0.63 $\pm$ 0.12). Both normal and treated rats of group II lost an average of 7 grams of body weight during the two week period.

It is concluded that while liver damage to the extent obtained in this investigation impairs the ability of the rat to metabolize even moderate quantities of fatty acids, minimal amounts are utilized as well as by the normal animal.

Total renal blood flow at any urine flow or extraction fraction. A. V. WOLF (introduced by E. F. Adolph). *Department of Physiology, University of Rochester School of Medicine, Rochester, N. Y.* (Read by title.)

While computing renal blood flows from arterio-venous differences of concentrations, it was noticed that current equations were inaccurate for substances of low extraction fraction, especially at rapid rates of urine flow. A paradox was evident for substances that are not extracted, where a negative flow would be computed. The usual equation for total renal blood flow (analogous to the Fick principle) has the form (1), $a = uU/(A\text{-}R)$, where a is rate of blood flow, u is rate of urine flow, U is concentration of a substance in the urine, and A and R are concentrations of this substance in arterial and renal vein blood, respectively. This relation becomes less exact as the urine flow increases and as the extraction fraction, $(A\text{-}R)/A$, decreases, because it recognizes no difference in rates of flow in renal artery and vein. The difficulties are avoided by the following practical method.

Let renal vein flow $r = (a\text{-}u)$. Then a is found by the equation $aA = (a\text{-}u)R + uU$ which states the law of conservation of matter for any substance, neglecting lymph flow. This gives (2), $a = u(U\text{-}R)/A\text{-}R)$. Here the value of a is unaffected by the factors of urine flow and extraction fraction, differing from equation (1) by the quantity $uR/(A\text{-}R)$, in which $R/(A\text{-}R)$ increases when the extraction fraction decreases. With substances of high extraction fraction like diodrast the advantage of the present equation is negligible. For inulin or urea, however, the errors avoided by its use are of the order of 4 to 14 per cent. For example, from data obtained for urea in a dog of Van Slyke *et al.* (Am. J. Physiol. **109:** 344, 1934) renal blood flow was computed by equation (1) as 167 cc./min. By the more general equation (2) a would be computed as 147 cc./min. The difference in computed flows amounts to 13.6 per cent.

Glucose reabsorption in the amphibian kidney. EARL H. WOOD[1] (introduced by A. N. Richards). *Laboratory of Pharmacology, University of Pennsylvania, Philadelphia.*

Previous studies have indicated that in the amphibian kidney blood glucose level is a determinant of glucose reabsorption by the kidney tubules. Shannon and Fisher have found that in the dog the capacity of the tubules to reabsorb glucose is independent of the blood level, glycosuria resulting when the glucose filtration rate exceeds the maximum tubular reabsorptive capacity

Since it seems unlikely that the glucose reabsorptive mechanisms of the amphibian and mammalian kidney are fundamentally unlike, an attempt has been made to learn more concerning this mechanism in the kidney of necturus.

In one series of experiments, fluid was collected simultaneously from a glomerulus and from the distal end of a proximal tubule, before and after injection of sufficient glucose to raise the blood glucose level above the kidney threshold (60 mgm. per cent). The calculated amount of glucose reabsorbed per 100 cc. of glomerular filtrate was as great or greater when blood glucose averaged 135 mgm. per cent (5 expts.) than when it averaged 44 mgm. per cent (10 expts.).

In 15 experiments after blockage of a glomerulus, the lumen of the proximal tubule originating from it was perfused with normal glomerular fluid, previously collected, while the blood glucose level was varied independently by intravenous injection of glucose. Average results follow:

NUMBER OF EXPERIMENTS	PERFUSION RATE, C.MM. PER HOUR		MGM. PER CENT GLUCOSE IN:			RATE OF GLUCOSE REABSORPTION, MGM. PER 1000 HOURS
	Injected fluid	Collected fluid	Plasma	Injected fluid	Collected fluid	
6	1.21(.96–1.43)	1.25(.96–1.52)	53(42–68)	50(35–64)	19(0–35)	.393(.195–.565)
9	1.01(.61–1.29)	1.03(.52–1.28)	143(86–190)	70(33–170)	29(3–115)	.374(.183–.640)

These results indicate that in necturi increase in blood glucose levels to 2–3 times that of the renal threshold does not diminish the rate of glucose reabsorption; they are in essential agreement therefore with the conclusions of Shannon and Fisher.

[1] National Research Council Fellow.

The effect of convulsive doses of metrazol on blood pressure: as employed therapeutically, during spinal anesthesia and during asthenia from curare. R. A. Woodbury, H. M. Cleckley (by invitation), Perry P. Volpitto (by invitation) and W. F. Hamilton. *Departments of Physiology and Pharmacology, Neuropsychiatry and Anesthesiology, University of Georgia, School of Medicine, Augusta.*

Optical records of pressure changes were obtained by the direct method, using the "hypodermic" manometer. In man systolic and diastolic arterial pressures are increased as much as 75 mm. Hg during the tonic convulsions and as much as 150 mm. Hg during the clonic convulsions. Records of the intra-abdominal pressure indicate that these elevated pressures are produced by contractions of the skeletal muscles. The increased intra-abdominal pressure supports and adds itself to the arterial pressure. In addition to this, the clonic contractions of the skeletal muscles have a pump-like action. This increases venous return and at times may push blood through the heart and into the pulmonary artery.

Immediately after the convulsions (usually one to one and one-half minutes after the metrazol injection) the systolic and diastolic arterial pressures are reduced, usually 20 to 25 mm. Hg below the normal values. Generally the pulse pressure is reduced and the heart is slowed. These pressure changes may result from extensive vasodilation which the heightened muscular activity would produce. Three to five minutes after the injection the arterial pressure became approximately normal.

Spinal anesthesia (level T-3) with procaine hydrochloride or asthenia produced by curare eliminates the initial rise in the blood pressure. This results from the limitation of the intensity and the extent of the muscular activity during the convulsions from metrazol. In the presence of asthenia from curare, the secondary fall of blood pressure is insignificant. During spinal anesthesia this secondary fall is pronounced and bradycardia and arhythmia are present. This dangerous reduction of blood pressure and bradycardia probably result from paralysis or paresis of the sympathetic nervous system (to the level T-3). This would allow excessive vagal stimulation by the metrazol.

In dogs and cats sub-convulsive doses of metrazol produce small changes in the blood pressure. Convulsive doses produce changes very much like those described above. Venous pressure records show that the convulsions markedly influence venous return to the heart.

The aid of Elkin Vogt while performing the preliminary experiments on dogs and cats is acknowledged.

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Topical projection of nerve fibers from local regions of the cochlea to the cerebral cortex of the cat. Clinton N. Woolsey and Edward M. Walzl (by invitation). *The Johns Hopkins University, School of Medicine, Baltimore, Md.*

By electrical stimulation of cochlear nerve fibers in the spiral osseous lamina near their terminations, it has been possible to obtain a point-to-point projection of the auditory nerve to the cerebral cortex of the cat. After exposure of the spiral osseous lamina by removal of the bony capsule of the cochlea together with the organ of Corti, fine steel electrodes, in-

sulated except at the tips, were placed on the free edge of the spiral osseous lamina at four points on the basal turn, at one on the second, and at one on the apical turn, (1, 3, 5, 7, 14 and 18 mm., respectively, from the basal end). When these points separately were stimulated with single condenser discharges, surface-positive potentials were evoked in rather restricted band-like regions extending dorso-ventrally through the auditory areas of both cerebral hemispheres.

It was found that the basal end of the cochlea projects to an area surrounding the superior end of the anterior ectosylvian sulcus, whereas the focal area for the apex was located just behind the superior end of the posterior ectosylvian sulcus. Intermediate regions of the cochlea were represented by intermediate zones in the auditory areas.

Corresponding points of ipsilateral and contralateral cochlae projected to the same cortical area of a single hemisphere and gave rise to potentials of similar magnitudes and latencies at particular cortical points.

Since the fibers of the cochlear nerve are distributed in an orderly manner to the organ of Corti, localized regions of which respond specifically to particular frequencies of the sound spectrum (Walzl and Bordley, these proceedings), the demonstration of a point-to-point projection of the fibers of the cochlear spiral to the cerebral cortex provides an anatomical basis for tonal localization in the cerebral mantle.

Temperature selection and the effect of temperature on movement in frog tadpoles. GRACE WORKMAN (by invitation) and KENNETH C. FISHER. *Department of Biology, University of Toronto, Toronto, Canada.*

When frog tadpoles are placed in a trough in which a temperature gradient exists, it is observed that as a consequence of their "random" movements they are more frequently observed in one temperature region than elsewhere. Frequency plots are readily made of the position of the tadpoles with respect to temperature. For this behaviour the term "temperature selection" will be used and "selected temperature" for the level of the peak of the curve (rather than the older terms "temperature preference" and "preferred temperature").

The selected temperatures for Rana pipiens, sylvatica and clamitans tadpoles raised at 20°C. are respectively 20, 17 and 27 ± 3°C. These values are only relatively significant since the selected temperature appears to vary with size and previous experience with respect to temperature.

In an attempt to discover the physiological basis of this behaviour Fisher and Elson have shown that for trout (Salvelinus fontinalis) and salmon (Salmo solar) the "selected temperature" is also the temperature of maximum movement. With this in mind measurements were made of the movement of *pipiens* tadpoles at constant temperature when stimulated with condenser discharges at a graded series of voltages. The distance moved was observed to increase with increasing stimulus strength up to a maximum and then to remain constant. From a series of such curves at several temperatures, the greatest movement at all strengths of stimulus was observed at about 20°C. Moreover, for individual tadpoles the selected temperature and the temperature of maximum movement change in a parallel fashion with changes in the temperature experience of the individuals.

It is therefore indicated that in frog tadpoles as well as in fish the selected temperature is also the temperature of maximum movement. This coincidence suggests that the physiological basis of the selection may be connected with the effect of temperature on the distance moved in response to stimulation.

Effects of milk diets on guinea pigs. ROSALIND WULZEN and ALICE M. BAHRS. *Department of Zoology, Oregon State College, Corvallis, and St. Helen's Junior College, Portland, Ore.*

Groups of young guinea pigs were fed rations of whole raw milk, pasteurized whole milk, raw skim milk and pasteurized skim milk. To the milks were added 10 per cent skim milk powder, adequate amounts of copper and iron, carotene and orange juice. The milk rations, straw and iodized salt were provided ad lib. Animals fed raw whole milk grew excellently and at autopsy showed no abnormality of any kind. Those on the pasteurized milk rations did not grow as well and developed a definite syndrome, the first sign of which was wrist stiffness. On pasteurized skim milk ration the syndrome increased in severity until the animals finally died in periods ranging from a month to a year or more. They showed great emaciation and weakness before death but remained in normal posture and had little tendency to paralysis of the limbs. Upon autopsy the muscles were found to be extremely atrophied and in most cases were streaked with closely packed, fine white lines of calcification running parallel to the muscle fibers. There were often lumps of tricalcium phosphate deposited under the skin, in the joint regions, between the ribs and indiscriminately in many body organs including heart and aorta.

It was found that raw cream given by mouth had power to cure the original wrist stiffness. An extract was made from raw cream which was able in a few days to restore the stiff wrists of affected animals to their normal limber condition. This active substance was found by Romeo Gouley to be methylvinylketone and was successfully synthesized by him. The synthetic product had active curative power.

When cod liver oil, ½ per cent, was substituted for carotene in the skim milk ration, in addition to stiffness the animals quickly developed paralysis. Their hind legs dragged and locomotion soon became impossible. We have found that synthetic methylvinylketone was able to restore normal locomotion to these animals provided the disease was not too far advanced.

The effect of gonadectomy upon the incidence of homoplastic adrenocortical transplants in rats. LELAND C. WYMAN and CAROLINE TUM SUDEN. *Physiological Laboratory of Boston University School of Medicine and the Evans Memorial, Massachusetts Memorial Hospitals, Boston, Mass.*

Although the "end-organ hormone" theory (developed to explain the rhythmicity of the sexual cycle, etc.) has been applied equally to both sexes, evidence is accumulating that while androgens inhibit pituitary activity, estrogens may stimulate it. Numerous investigators agree that the injection of androgens is followed by decrease in the weights of the pituitary and adrenal glands, but that the injection of estrogens is followed by an increase; and that gonadectomy is followed by an increase in the

weight of the two glands in males, but by no change or a decrease in females.

It has been established that the growth of transplanted adrenocortical tissue depends upon pituitary activity (presumably adrenotropin production). Intramuscular autoplastic transplants are successful in about 95 per cent of cases, but non-sibling homoplastic transplants are successful in about 30 per cent. The effect of gonadectomy upon this latter incidence was therefore studied.

Young mature rats were used. Dorsal intramuscular homoplastic transplants of adrenal cortex were made in adrenalectomized nonsibling recipients. Gonadectomy was performed simultaneously. Successful transplantation was judged by health, survival and examination of the grafts at biopsy, 60 to 125 days after operation. Successful grafts were obtained in 11 of 38 male (28.9 per cent) and in 21 of 65 female (32.3 per cent) non-gonadectomized controls. The incidence in the gonadectomized rats was 21 of 39 males (53.8 per cent) and 10 of 35 females (28.5 per cent). The increased incidence in males and lack of significant change in females, confirms the opinions stated above.

There is not complete agreement as to the duration of pituitary changes following gonadectomy. Some say that they persist for years. In 10 male rats castrated two weeks before adrenal homotransplantation, 4 (40 per cent) grafts were successful; and in 45 males castrated 2 to 3 months before homotransplantation, 12 grafts were successful (26.6 per cent), a figure not significantly different from that of the controls. Apparently the changes responsible for the increased incidence of successful grafts (53.8 per cent) when castration and transplantation are simultaneous do not persist indefinitely.

Micromanipulative studies on vascular responses remote from a traumatized region. BENJAMIN W. ZWEIFACH (introduced by Robert Chambers). *Washington Square College, New York University, New York City.*

The effects of trauma, to tissues remote from the area under observation, were followed in the tongue, mesentery and intestinal wall of the frog. Here, the significant initial alteration occurs in the arterioles. This is in contrast to the observed responses in areas injured with microneedles, where the venules and capillaries are primarily involved. Of the two controlling factors regulating the caliber of the small arterioles, the constrictor influence is primarily affected. This is indicated by the fact that the arterioles become refractile to constrictor nervous influences. They remain partially dilated and are no longer observed to narrow down, but still respond to local mechanical stimulation. The capillary circulation, instead of being confined to a fraction of the available channels, is now distributed throughout the capillary bed. As a result a considerable portion of the blood remains in the tissues.

The persistent opening of progressively increasing numbers of capillaries produces a gradual slowing of the venous flow. The larger arteries then become considerably narrowed. This serves only to slow down the widely distributed blood flow in the capillary bed. As a result the blood which reaches the venules gradually becomes more venous in character. The